FIBRINOGEN CONCENTRATE SAFETY FOR THE CORRECTION OF HYPOFIBRINOGENEMIA IN COAGULOPATHIC SURGICAL PATIENTS

ALZIRA MANUELA DA ROCHA GOMES VIEGAS

Doctoral Programme in Medicine, in the field of Clinical Research and specialty in Immunohemotherapy, at Faculdade de Ciências Médicas | NOVA Medical School of NOVA University Lisbon

December, 2024

FIBRINOGEN CONCENTRATE SAFETY FOR THE CORRECTION OF HYPOFIBRINOGENEMIA IN COAGULOPATHIC SURGICAL PATIENTS

Alzira Manuela Da Rocha Gomes Viegas

Supervisor: Jorge Natalino Ramos Lima, MD, PhD, Invited Assistant Professor at Faculdade de Ciências Médicas | NOVA Medical School of NOVA University Lisbon

A thesis submitted in fulfillment of the requirements for the Doctoral Degree in Medicine, in the field of Clinical Research with specialization in immunohemotherapy

December, 2024

Preface

All scientific research garners the interest and effort of researchers for a reason, whether it leads to great or small advances. It is not feasible to believe that it does not represent the outcome of a collective contribution.

This work is no exception and results from a long-standing struggle to demonstrate a single clinical point, which also lends its name to this thesis. The journey began nearly two decades ago at Hospital de Santa Cruz - Carnaxide, in the perioperative period of adult and pediatric cardiac surgeries. During this time, patients were experiencing significant bleeding, whether in the operating theatre or the Intensive Care Unit. The doctors from the Transfusion Medicine Department would rush to the scene and remain with their colleagues until a resolution was reached, tirelessly working and hoping that the bleeding would cease. Each patient presented us with a unique struggle, regardless of whether they were elderly, middle-aged, young, or newborn. As a team, we gave our utmost effort to save their lives. However, there were instances where, despite utilizing every therapeutic tool at our disposal, we could not stop the bleeding. My sentiment was that we could improve and expedite things, only, at the time, I was not sure how (given that we were in the early 2000s).

Most protocols addressed major bleeding associated with the perioperative period, obstetrics, and trauma in the same way. These protocols, both past and present, are fundamentally based on therapeutic approaches grounded in war scenarios. They utilize what we term “cell ratios” of red blood cells to restore hemoglobin, and fresh frozen plasma to replenish lost coagulation factors and platelets. The rationale underpinning this cell ratio therapy essentially aims to replace the whole blood lost by the patient during this process.

We also had another type of blood component, which at that time was considered the last frontier in trying to halt severe bleeding. This was cryoprecipitate, which contains fibrinogen, FXIII, FVIII, von Willebrand factor, and fibronectin. However, most of the time, when we resorted to this option, the outcome was typically poor: severe morbidity and, more often than not, a high mortality rate. During that period, other colleagues encountered similar issues, and a small group of dedicated clinicians in our country initiated a multidisciplinary approach to a field known as acquired coagulopathies.

We began to tackle acquired coagulopathies in a more targeted manner, following the international scientific findings that had emerged by then and are now well-established. These findings demonstrated that within each patient, pathophysiological mechanisms follow sequential steps, adapting of course to patients’ unique characteristics and specific clinical scenarios. Besides red blood cells, conditions such as hyperfibrinolysis (premature destruction of the clot), hypothermia, hypocalcemia, and acidosis could undermine clot stability. Moreover,

coagulation factors and inhibitors are not required simultaneously in the same manner or dosage; rather, they are needed step by step in a more targeted therapy.
So, some of us started to use *point-of-care devices* to study hemostasis at the patient's bedside and began directing therapy focused on the specific needs of each patient at any given moment. This is because, in cases of major bleeding, things can change quite swiftly – within a matter of minutes.

Instead of plasma, coagulation factor concentrates such as fibrinogen concentrate, and pharmacological agents like tranexamic acid (used to control hyperfibrinolysis), have become our first choice. This is based on both laboratory results and clinical outcomes. This work was indeed exciting, originating from Portuguese Institutions. However, the reality is that efforts to change mindsets, and in doing so, enhance clinical outcomes for patients, are not always straightforward. They unquestionably require time and patience. Such was the case in our situation, but today, we can handle acquired coagulopathies in a bleeding patient far more effectively and differently than we did 20 years ago.

In this vast and complex field of coagulation, which many health professionals deem tedious, this work may seem insignificant. However, after many years of dealing with bleeding patients, I view it as quite important because it offers a new approach to blood treatment. This method is goal-directed and safer, striving to understand each situation within a comprehensive model. The aim is to determine what can be done in an extensive strategy to minimize mortality and morbidity and to conserve the patient's own blood.

When considering the administration of a drug in a novel clinical scenario, efficacy and safety are paramount. This goal-oriented approach repurposes traditional substances, such as coagulation factor concentrates, for new applications. This has sparked a substantial number of studies, particularly regarding fibrinogen concentrate. Fibrinogen concentrate has been used for several decades worldwide, in both children and adults with congenital deficiencies. It boasts an excellent safety profile and consistently yields good results. However, in this case, we are administering it to patients with transitory fibrinogen deficiency. As a procoagulant, in clinical situations where a delicate balance exists between thrombosis and the urgent need to achieve hemostasis, safety is of the utmost importance.

This thesis is the result of what was exposed above and focuses only on one aspect – fibrinogen concentrate's safety for bleeding patients in the perioperative period.To achieve this objective, two scientific studies were conducted and published in 2024, though the underlying fieldwork was initiated long before.

- **Gomes M**, Ângelo-Dias M, Duarte GS, Dias SS, Serra SS, Lima J. Safety of Fibrinogen Concentrate in Non-Trauma and Non-Obstetric Adult Patients during Perioperative Care:

Systematic Review and Meta-Analysis. J Clin Med. 2024 Jun 14;13(12):3482. doi: 10.3390/jcm13123482. PMID: 38930009; PMCID: PMC11204778.
Impact Factor: 3.0; Q1

- **Gomes M**, Ângelo-Dias M, Lima J. Safety of Fibrinogen Concentrate for Correcting Perioperative Bleeding-Associated Hypofibrinogenemia in Adults: A Single-Center Experience. J Clin Med. 2024 Oct 9;13(19):6018. doi: 10.3390/jcm13196018. PMID: 39408077; PMCID: PMC11477569.
  Impact Factor: 3.0; Q1

This PhD research project was approved by Comissão de Ética da NOVA Medical School/Faculdade de Ciências Médicas - UNL (N° 14/2022/CEFCM) and by Comissão de Ética para a Saúde do Hospital da Luz (N° CES/58/2022/ME).

To twenty-six children who were born with a cardiac disease. They all fought for their lives when they were too young and too much innocent to understand what was happening. While some of them won the battle, others did not. They were a true inspiration to me and represented a line mark in my professional and personal live.

To my parents as they always were there for me and showed me the right way to follow. I will always miss them and hope to keep up with the high standards that they set for my life.

Acknowledgments

First, I sincerely wish to thank my supervisor, Professor Jorge Lima, for guiding me, always believing in my work, and providing me strength during moments of doubt. I firmly believe that without his guidance, this thesis would not exist.

I am also grateful to my colleagues at Santa Cruz Hospital and Luz Lisboa Hospital, for their unwavering faith in my scientific reasoning and their unquestioning acceptance of my new therapeutic decisions.

There are so many names to mention that I would not dare to write them all, for fear of leaving anyone out. To Dr. Francisco Soares and Dr. Manuela Soares (Hemovida), thank you for consistently supporting my clinical opinions, acknowledging my need to introduce new ideas, and providing me the opportunity to cultivate the field that I am most passionate about.
To all the technicians with whom I work daily, thank you for accepting me, never questioning my decisions, patiently tolerating my in-depth scientific explanations, and being the steadfast pillars amidst our often challenging day-to-day tasks.
A special thank you to Dr. Ana Cristina Silva and Dr. Pedro Gomes for their assistance in data collection for the prospective study at Hospital da Luz Lisboa.
To Dr. Miguel Ângelo Dias, for his assistance and encouragement in the production of both papers that serve as the scientific foundation of this thesis.

Finally, I extend my gratitude to my husband for his extraordinary patience and support towards my work, which he knows is my passion. Additionally, a special thanks to both my sons for their curiosity, support, and interest in my thesis.

Resumo

O fibrinogénio plasmático é um fator da coagulação que desempenha um papel crítico na hemostase primária e secundária, uma vez que constitui a base do coágulo sanguíneo, após lesão de um vaso. Trata-se de uma molécula grande, estruturalmente intrincada e que circula em altas concentrações no plasma.

A hemorragia leva à perda, diluição e consumo de fibrinogénio, enquanto que a hipotermia e a acidose alteram a sua capacidade funcional. Em qualquer doente que apresente uma hemorragia aguda, os níveis de fibrinogénio dependem de um equilíbrio delicado entre a sua síntese, consumo e degradação. Os procedimentos cirúrgicos, em particular as cirurgias major, estão frequentemente associadas a hemorragia, e esse facto pode representar um desafio para a hemostase. Um número crescente de estudos sustenta o facto de que a reposição de fibrinogénio ajuda a melhorar a coagulopatia adquirida e minimizar a necessidade de transfusão sanguínea, que não é inócua, além do sangue ser um bem escasso, nem sempre disponível e indiscutivelmente caro. Dada a sua importância e papel único na manutenção da hemostase, a sua rápida suplementação torna-se essencial para alcançar o controlo das perdas sanguíneas, uma vez que o aumento da síntese de fibrinogénio, pode não ser suficiente para compensar o seu consumo.

Existem três possibilidades de suplementação de fibrinogénio: plasma, crioprecipitado e concentrado de fibrinogénio. O plasma e o crioprecipitado não são a fonte ideal para reposição de fibrinogénio, enquanto que o concentrado de fibrinogénio tem a vantagem de possuir uma quantidade fixa de fibrinogénio num pequeno volume, ser submetido a pasteurização e inativação viral e estar rapidamente disponível. No entanto, as evidências disponíveis sobre a relação benefício/risco do uso de concentrado de fibrinogénio para diminuir a hemorragia e complicações relacionadas, em diferentes situações clínicas, não são amplamente aceites. A maioria dos ensaios clínicos randomizados disponíveis e outros estudos clínicos concluem que o concentrado de fibrinogénio mostra eficácia e parece ser seguro, mas persistem dúvidas e resultados discrepantes. Várias décadas de farmacovigilância evidenciam poucos relatos de eventos adversos, mas devemos considerar que, além do facto de o concentrado de fibrinogénio ser um pró-coagulante, ele é administrado a doentes com coagulopatias e com outros fatores de risco cumulativos para o desenvolvimento de eventos trombóticos/tromboembólicos.

O objetivo deste projeto foi avaliar a segurança do concentrado de fibrinogénio em doentes com coagulopatias adquiridas aos quais foi administrado concentrado de fibrinogénio, para correção de hipofibrinogenémia, tendo como foco a mortalidade e eventos trombóticos/tromboembólicos diagnosticados por métodos clínicos, laboratoriais e radiológicos.

Esta tese foi baseada em dois estudos: uma revisão sistemática com meta-análise sobre a segurança do concentrado de fibrinogénio em doentes adultos durante o período peri operatório, e um estudo observacional prospetivo realizado num só centro, que incluiu todos os doentes

adultos submetidos a cirurgia programada ou de emergência com coagulopatia hemorrágica associada a administração de concentrado de fibrinogénio, dentro dos limites terapêuticos para este efeito, com a intenção de corrigir a hipofibrinogenémia.

No primeiro estudo, foi realizada uma pesquisa bibliográfica extensa no *PubMed/Medline*, *EMBASE, Scopus, Web of Science, Cochrane Database of Systematic Reviews* e *Cochrane Central Register of Controlled Trials*. Dois revisores administraram o processo de forma independente, examinando meticulosamente os estudos recuperados, extraindo dados e avaliando a integridade metodológica. Um terceiro revisor resolveu qualquer desacordo entre eles.

O segundo estudo foi realizado no Hospital da Luz Lisboa, com o objetivo de reproduzir no mundo real, a abordagem à hemorragia associada à hipofibrinogenémia, tendo os doentes sido acompanhados até à data da alta hospitalar. Os dados colhidos foram abrangentes, incluindo idade, sexo, tipo de cirurgia, comorbidades associadas, terapia anticoagulante e/ou antiagregante e número de componentes sanguíneos transfundidos. Os dados laboratoriais sobre concentração plasmática de fibrinogénio, resultados de testes viscoelásticos, hemoglobina e contagem de plaquetas antes e após a cirurgia também foram colhidos. Os objetivos primários foram a taxa de mortalidade à data de alta e quaisquer eventos trombóticos ou tromboembólicos relatados, incluindo trombose venosa profunda, embolia pulmonar e enfarte do miocárdio até a alta hospitalar. No início do estudo, fatores de risco adicionais para eventos trombóticos/tromboembólicos, como: obesidade, tabagismo, história familiar e pessoal pregressa de eventos tromboembólicos, trombofilia hereditária, imobilidade, dias de repouso no leito e medicamentos, como anticoncecionais orais, terapia de reposição hormonal e glicocorticoides, também foram colhidos. Uma vez que estes podem representar variáveis confundentes, esses dados permitiram uma análise de estratificação estatística posterior.

A revisão sistemática com meta-análise englobou dez estudos envolvendo 1391 pacientes. Verificou-se uma diminuição do risco de eventos tromboembólicos totais em doentes tratados com fibrinogénio em comparação com o grupo controlo (OR 0,65; IC 95% 0,43 a 0,98, I^2 = 0%). Além disso, quando o fibrinogénio foi usado profilaticamente, resultou em menor tempo de permanência na Unidade de Cuidados Intensivos (MD -1,50; IC 95% -2,64 a -0,36), quando comparado ao seu uso terapêutico. No entanto estes resultados devem ser analisados com alguma cautela. A análise de sensibilidade em estudos de cirurgia cardiovascular não revelou diferenças estatisticamente significativas.

O segundo estudo, que abrangeu 91 doentes cirúrgicos, e onde se verificou a morte de oito doentes, não revelou nenhuma associação percetível entre a administração de concentrado de fibrinogénio e mortalidade. Da mesma forma, oito doentes apresentaram eventos tromboembólicos após a cirurgia, mas todos se encontravam vivos à data da alta hospitalar. Ao contrário de outros relatos, houve uma distribuição igualitária de sexos entre esses doentes.

Variáveis confundentes, como doenças concomitantes e história pessoal ou familiar de doença trombótica, podem fornecer explicações alternativas para esses eventos tromboembólicos. Desses oito doentes, apenas um não apresentava comorbidades associadas. Entre os sete restantes, cinco tinham história pessoal de eventos tromboembólicos e um tinha história familiar positiva. Estes doentes também apresentaram diversas situações clínicas, como infeção por COVID-19, neoplasias e fibrilhação auricular. Os doentes que sofreram eventos tromboembólicos durante a fase de acompanhamento, também tiveram tempos de internamento hospitalares mais longos e receberam mais unidades de concentrado eritrócitário.

Os nossos dados indicam que a idade, a incidência de eventos tromboembólicos prévios e as cirurgias de emergência, se associaram a uma maior necessidade de transfusão de componentes sanguíneos e períodos de internamento hospitalar mais longos. Embora o uso de concentrado de fibrinogénio não pareça aumentar o risco de eventos tromboembólicos, os doentes que necessitaram de doses mais altas de concentrado de fibrinogênio também receberam mais unidades de eritrócitos. Esse achado está alinhado com uma revisão sistemática que analisou a adminstração de concentrado de fibrinogénio para controlo da hemorragia em situações de emergência.

Os resultados de ambos os estudos estão alinhados e parecem reforçar as mesmas conclusões. Por um lado, os resultados da revisão sistemática com meta-análise indicam que o uso de concentrado de fibrinogénio no período peri operatório de doentes adultos sem coagulopatia traumática e não obstétricos, pode levar a benefícios potenciais. Por outro lado, os resultados do estudo prospetivo indicam um perfil de segurança favorável para o concentrado de fibrinogénio em doentes cirúrgicos, demonstrado por uma baixa incidência de óbitos e eventos tromboembólicos, atribuídos a outros fatores. No entanto, pesquisas futuras devem priorizar a minimização de viés e o aumento da robustez estatística para esclarecer de forma mais eficaz as medidas de segurança do doente, que possam ser clinicamente significativas.

Abstract

Fibrinogen, a clotting factor, plays a critical role in both primary and secondary hemostasis, forming the basis of the clot. This structurally intricate, large molecule circulates at high concentrations.

Bleeding can lead to fibrinogen loss, dilution, and consumption, while hypothermia and acidosis impair its function. The fibrinogen levels in any patient presenting with an acute hemorrhage are dependent on a delicate balance between synthesis, consumption, and degradation. Surgery is often associated with bleeding, which can pose a challenge to hemostasis. An increasing number of studies support the concept that fibrinogen replacement can improve acquired coagulopathy and reduce the need for blood transfusions. These transfusions are not innocuous and, due to their scarcity and significant cost, may not always be available.

Given its importance and unique role in maintaining hemostasis, rapid fibrinogen supplementation becomes essential to achieve blood loss control, since an increase in synthesis alone may not compensate for consumption.

There are three possibilities for fibrinogen supplementation: plasma, cryoprecipitate, and fibrinogen concentrate. Plasma and cryoprecipitate have not been proven to be the ideal sources for fibrinogen replacement. Meanwhile, fibrinogen concentrate appears to have advantages as it has a fixed amount of fibrinogen in a small volume, undergoes pasteurization and viral inactivation, and is rapidly available. However, the existing evidence on the benefit/risk ratio of using fibrinogen concentrate to decrease bleeding and related complications in different clinical situations is not widely accepted. The majority of available randomized clinical trials and other clinical studies conclude that fibrinogen concentrate is effective and appears safe, but there are some conflicting data. Several decades of pharmacovigilance have reported relatively few adverse events. However, we must take into account that fibrinogen concentrate is a procoagulant that is administered to coagulopathic patients, who often have other accumulated risk factors for developing a thrombotic/thromboembolic event.

The goal of this project was to evaluate the safety of fibrinogen concentrate in coagulopathic patients who received it to rectify hypofibrinogenemia. The focus was on studying mortality and thrombotic/thromboembolic events diagnosed by clinical, laboratory, and radiological methods.

This thesis was based on two studies: a systematic review with a meta-analysis on the safety of fibrinogen concentrate in non-trauma, non-obstetric adult patients during the perioperative period, and a single-center, prospective observational study. The latter included all adult patients undergoing scheduled or emergency surgery, presenting with bleeding coagulopathy, and receiving administration of fibrinogen concentrate within therapeutic ranges for the correction of hypofibrinogenemia.

In the initial study, an exhaustive online search was performed on PubMed/Medline, EMBASE, Scopus, Web of Science, Cochrane Database of Systematic Reviews, and the Cochrane Central Register of Controlled Trials. Two reviewers independently undertook the process, carefully screening the retrieved studies, extracting data, and assessing methodological integrity. Any discrepancies between them were resolved by a third-party reviewer.

The second study was conducted at Hospital da Luz Lisboa, to encapsulate a real-world approach to bleeding related to hypofibrinogenemia. Patients were followed until their discharge from the institution. Comprehensive data were collected, which included age, sex, type of surgery, associated comorbidities, anticoagulant and/or anti-aggregating therapy, and the number of blood transfusions. Laboratory data – such as plasmatic fibrinogen concentration, *point-of-care* results, hemoglobin, and platelet count before and after surgery – were also collected. The primary outcomes were the mortality rate at discharge and any reported thrombotic or thromboembolic events, including deep vein thrombosis, pulmonary embolism, and myocardial infarction until the point of hospital discharge. At baseline, additional risk factors for thrombotic/thromboembolic events, such as obesity, smoking, family and past personal history of venous thromboembolism(VTE), inherited thrombophilia, immobility, days of bed rest, and drugs like oral contraceptives, hormone replacement therapy, and glucocorticoids, were also collected. Given that these could be confounding variables, this data set allowed for further statistical stratification analysis.

The systematic review, which included a meta-analysis, covered ten studies involving 1,391 patients. It revealed a decreased risk of total thromboembolic events in patients treated with fibrinogen, compared to the control group (OR 0.65, 95% CI from 0.43 to 0.98, with I^2 at 0%). Additionally, when fibrinogen was used prophylactically, it led to reduced lengths of intensive care unit (ICU) stays (MD -1.50, 95% CI from -2.64 to -0.36) compared to its therapeutic use. A sensitivity analysis focusing on cardiovascular surgery studies found no statistically significant variation.

The second study, which included 91 surgical patients, revealed no conspicuous associations between the administration of fibrinogen concentrate and mortality among the eight patients who died. Likewise, eight patients encountered thromboembolic events after surgery, but all were alive at the time of hospital discharge. In contrast to other reports, the distribution of genders among these patients was evenly balanced.

Confounding variables, such as concurrent illnesses and personal or familial history of thrombotic disease, could provide alternate explanations for these thromboembolic events. Among these eight patients, only one had no associated comorbidities. Of the remaining seven, one had a positive family history of thrombotic disease. These patients also presented with varied conditions, which included prior thromboembolic events in two cases, a history of COVID-19 infection, cancer, and atrial fibrillation in the others. Patients who experienced thromboembolic

events during the follow-up phase also had longer hospital stays and received more units of red blood cells (RBCs).

Our findings suggest that factors such as age, the incidence of thromboembolic events, and emergency surgeries were associated with a greater need for blood transfusions and extended hospital stays. Although the application of fibrinogen concentrate did not appear to increase the risk of thromboembolic events, patients who required higher doses of fibrinogen concentrate also received a greater quantity of RBC units. This observation is consistent with a systematic review that investigated the employment of fibrinogen concentrate in addressing hemorrhaging during emergencies.

The results from both studies are aligned and seem to strengthen the same conclusions. On the one hand, the systematic review with meta-analysis indicates that the use of fibrinogen concentrate in the perioperative care of non-trauma and non-obstetric adult patients may lead to potential benefits. On the other hand, the prospective study demonstrates a favorable safety profile for fibrinogen concentrate in surgical patients, as indicated by a low incidence of deaths and thromboembolic events, primarily attributed to other factors. However, future research should prioritize minimizing bias and enhancing statistical robustness to more effectively highlight clinically significant patient safety measures.

Table of Contents

List of figures

Chapter I

Chapter III

Chapter IV

List of Tables

Chapter III

Chapter IV

Thesis Outline

Chapter I provides a comprehensive introduction to the current understanding of endometriosis and the immune system, investigating the linkage between the two, as well as exploring the correlations between immune evasion in cancer and endometriosis.

Chapter II presents the main objectives of this thesis.

The subsequent chapters present the two studies on which this PhD thesis is based. Each chapter contains the content of a published manuscript, subdivided into the main sections of abstract, introduction, materials and methods, results, discussion, and conclusion.

Chapter III presents a systematic review with meta-analysis on the safety of fibrinogen concentrate in non-trauma and non-obstetric adult patients during perioperative care.

- **Gomes M**, Ângelo-Dias M, Duarte GS, Dias SS, Serra SS, Lima J. Safety of Fibrinogen Concentrate in Non-Trauma and Non-Obstetric Adult Patients during Perioperative Care: Systematic Review and Meta-Analysis. J Clin Med. 2024 Jun 14;13(12):3482. doi: 10.3390/jcm13123482. PMID: 38930009; PMCID: PMC11204778.

Chapter IV describes a single-center experience and safety analysis of using fibrinogen concentrate to correct perioperative bleeding-associated hypofibrinogenemia in adults.

- **Gomes M**, Ângelo-Dias M, Lima J. Safety of Fibrinogen Concentrate for Correcting Perioperative Bleeding-Associated Hypofibrinogenemia in Adults: A Single-Center Experience. J Clin Med. 2024 Oct 9;13(19):6018. doi: 10.3390/jcm13196018. PMID: 39408077; PMCID: PMC11477569.

Chapter V presents a comprehensive discussion of this work, including its strengths and limitations, final remarks, and future perspectives.

CHAPTER I

General Introduction

1 Overview of Fibrinogen in modern clinical practice

In today's fast-paced world, we are driven by the pursuit of novelty. Technology, fashion, speed, social media, and the sway of trends dominate our attention. "New" is the characteristic feature of value, while "old" is frequently dismissed as irrelevant. Regrettably, this mindset extends beyond consumer culture – it affects how we view knowledge and even people. In our eagerness to embrace innovation, we often overlook valuable elements from the past, including critical discoveries in science and medicine.

Fibrinogen, a clotting factor and a remarkable molecule with astounding functions, embodies this predicament. Although it is vital for life, its long-standing discovery has resulted in widespread neglect in both research and clinical focus. However, without fibrinogen, a modest wound or blood vessel injury could lead to fatal, uncontrolled bleeding. Thankfully, just like numerous other issues within our organism, science has devised alternatives to tackle clotting complications; nevertheless, fibrinogen itself remains irreplaceable.

The paradox is clear: fibrinogen is both indispensable and undervalued. Science has acknowledged its existence for centuries and it has been studied extensively. However, its very familiarity has rendered it "old news" in the eyes of many. Sadly, the truth is that we do not understand it as well as we assume. Recent discoveries have exposed gaps in our knowledge, proving that there's still much to learn about its mechanisms and potential applications.

Modern medicine heavily relies on blood transfusions, a resource that is scarce and not replicable in a laboratory. It depends on the goodwill and civic sense of blood donors. Blood transfusions are also costly, imposing a financial burden on healthcare systems. Consequently, there is a growing emphasis on the concept of Patient Blood Management (PBM), a clinical approach that aims to minimize blood loss, optimize its use, and centralize decisions concerning the patient's own blood. Fibrinogen assumes a pivotal role in PBM, especially during the perioperative period when the demand for blood transfusions peaks. By fortifying the body's natural clotting processes, fibrinogen reduces the dependence on donor blood, making it an indispensable tool in addressing both the medical and logistical challenges in healthcare. Despite its significance, however, fibrinogen has diminished in focus in clinical practice.

This work aims to bring fibrinogen back into focus, advocating for its integration into daily medical decisions. Rediscovering its potential is not simply a matter of scientific curiosity; it is essential for improving patient outcomes and optimizing the use of valuable blood resources.

2 Hemostasis: Mechanisms of coagulation

2.1 The Y-shaped cascade

After decades of research aimed at understanding hemostatic mechanisms, various groups of investigators proposed the Y-shaped cascade model of coagulation in the 1960s. In this model, enzymes cleave proenzymes to generate the subsequent enzyme in the cascade (Figure 1). Ultimately, a fibrin network is formed, which leads to the generation of a stable hemostatic clot [1].

The model is divided into two pathways: the extrinsic and intrinsic pathways, which converge in the common pathway. Both pathways activate Factor X to Factor Xa. This, in turn, activates prothrombin to thrombin, which subsequently cleaves fibrinogen to form fibrin. The cascade model is notably helpful as it provides insight into the functionality of coagulation enzymatic steps and elucidates the role of each coagulation factor.

However, this model only explains what occurs in a static system where the fluid does not interact with the vascular wall or cell surfaces reasonably. It also suggests that the extrinsic and intrinsic pathways operate as independent and redundant methods, even though clinical manifestations in patients with congenital factor deficiencies contradict this concept [2].

Therefore, this model does not accurately portray what truly happens *in vivo*. For example, patients suffering from factor XII deficiency do not exhibit a bleeding tendency, even though they often present with a significantly prolonged activated partial thromboplastin time (aPTT), suggesting a disruption in the intrinsic pathway. In contrast, deficiencies in factor XI might be linked to a bleeding tendency, even though the prolongation of aPTT does not consistently align with the extent of anticipated hemorrhage, which is typically less severe than that seen in hemophilia. Hemophilia presents an intriguing scenario, as patients experience severe bleeding episodes, despite having a normal prothrombin time (PT), a measure of the extrinsic pathway activity, and a prolonged aPTT. This raises crucial questions: Why does the extrinsic pathway fail to make up for the impairment in the intrinsic pathway? Why do hemophiliacs bleed despite the ostensible independence of the extrinsic system [2]?

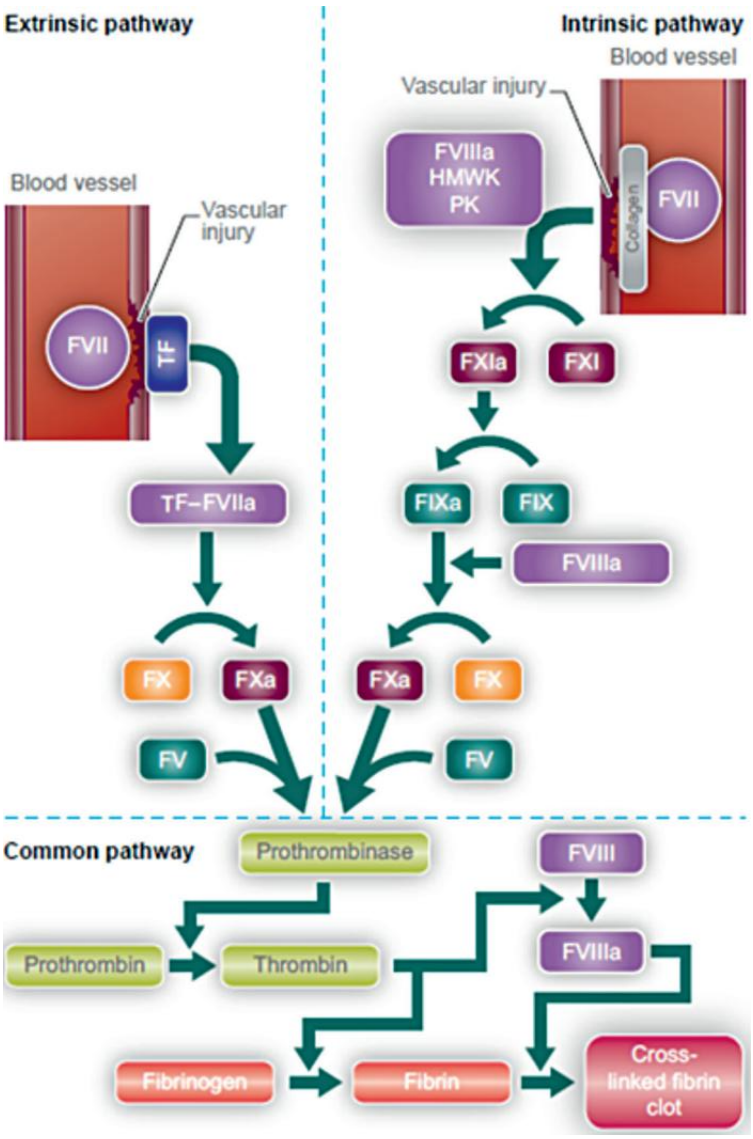

Figure 1 - The coagulation cascade in its classic Y-shaped form. Adapted from Gomes M, 2014 [3].

2.2 The cell-based model of Coagulation

The cell-based model of coagulation provides a more accurate portrayal of hemostasis, emphasizing the crucial role that cell surfaces play in coagulation. Specifically, two types of cells are crucial to this process: Tissue Factor (TF)-bearing cells, and platelets [2]. The velocity and efficacy of coagulation reactions are significantly impacted by the presence of viable cell membranes, which facilitate the interactions required for thrombin generation [4]. Thrombin, which is vital for clot formation, is directly generated on the activated platelet surface, while both the intrinsic and extrinsic pathways concurrently produce Factor Xa. According to this model, coagulation unfolds in four phases: initiation, amplification, propagation, and stabilization. The primary hemostasis, intrinsic, and extrinsic pathways occur simultaneously (Figure 2).

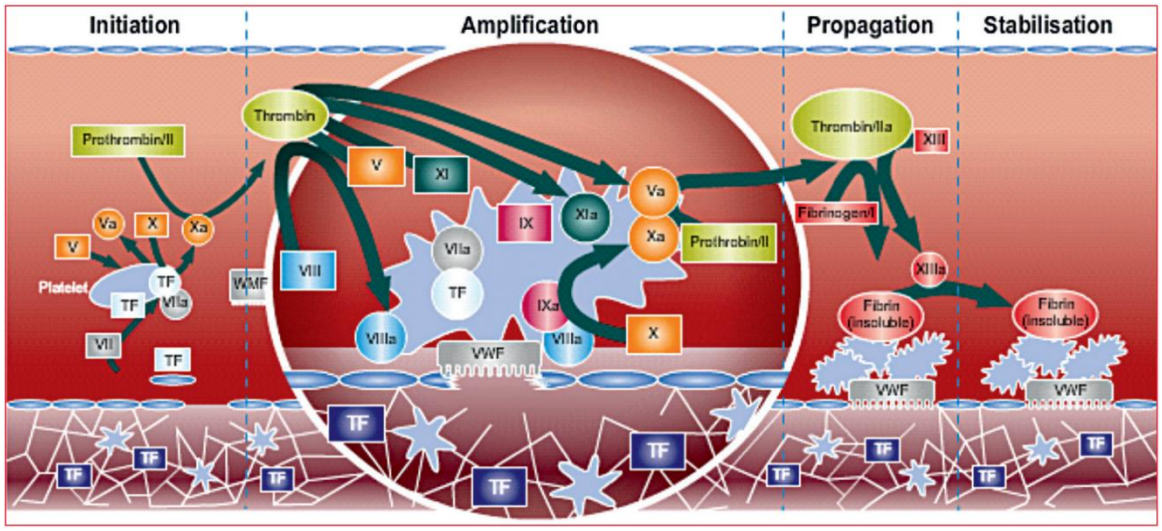

Figure 2 - The cell-based model of coagulation. Adapted from Gomes M, 2014 [3].

2.2.1 Initiation phase

Coagulation is initiated on a TF-bearing cell, which forms a complex with small amounts of FVIIa (TF/FVIIa). This process, often referred to as the extrinsic pathway, derives its name from the fact that these TF-bearing cells are normally located outside the vascular system—thus, they are "extrinsic" to the blood. TF-expressing cells include stromal fibroblasts, mononuclear cells, macrophages, and endothelial cells. Under normal conditions, TF is not in contact with blood; however, injury or inflammation can expose TF to the bloodstream, triggering coagulation. Interestingly, even in healthy individuals, the TF pathway is active at low levels, generating small amounts of activated coagulation factors. While this leads to low levels of thrombin, it does not cause clot formation unless there is vascular injury [2, 4].

2.2.2 Amplification phase

Following vascular injury, certain components typically confined to the vasculature due to their large size—such as platelets, FVIII, and von Willebrand factor (vWF)—are exposed to the blood flow. These factors come into contact with thrombin, which is generated on the surface of the TF-bearing cells. Platelets adhere to the vessel wall via vWF, which binds to collagen in the vessel and a specific receptor on the platelets. After adhesion, platelets become activated, causing them to change shape, degranulate, and spread over the damaged endothelium. This process recruits additional platelets, promoting further aggregation and degranulation [2].

Thrombin plays a crucial role in the activation of coagulation factors, such as FV, FVIII (cleaved from vWF), and possibly FXI. Notably, this might explain why FXII, along with other

contact factors, is not always essential for coagulation. While the small amount of thrombin generated during the amplification phase cannot form a clot by itself, it significantly contributes to the subsequent propagation phase by activating additional factors and platelets [1, 2, 4].

2.2.3 Propagation phase

In the propagation phase, the tenase complex (FIXa/FVIIIa) activates FX to become FXa, which then combines with FVa to form the prothrombinase complex. This complex converts substantial amounts of prothrombin into thrombin, resulting in a thrombin surge. This substantial increase in thrombin production leads to the cleavage of fibrinogen into fibrin monomers, which then polymerize and assist in solidifying the initial platelet plug into a stable fibrin clot [2].

In hemophilia, the propagation phase is impaired. This is because patients lack functional FVIII or FIX, which are necessary for the proper formation of the tenase complex. Additionally, the movement of FXa from the TF-bearing cell surface to the platelet surface is hindered by two potent plasma inhibitors: Tissue Factor Pathway Inhibitor (TFPI) and antithrombin. These inhibitors rapidly remove FXa from circulation, preventing its activity for an extended period. As a result, FXa has a very short half-life (less than 1 minute) in hemophilia patients, preventing the necessary burst in thrombin for clot formation [2].

2.2.4 Stabilization phase

The stabilization phase entails the cross-linking of fibrin by FXIII, which produces a mechanically stable clot. This stage is critical for wound healing because it consolidates the clot, halting further bleeding and allowing tissue repair to commence. Many factors, such as calcium concentration, pH, and platelet count, influence clot stability. Moreover, the diameters of the fibrin fibers and the geometry of the fibrin network contribute significantly to the overall strength of the clot.

Higher concentrations of thrombin are conducive to the creation of more stable clots, while lower concentrations are more likely to produce fragile clots that are more susceptible to fibrinolysis and subsequent bleeding. On the other hand, more resilient clots can encourage thrombosis. Variants of fibrinogen (for instance, γ´ fibrinogen) can lead to thinner fibrin fibers that exhibit increased branching, affecting clot formation. Thus, the formation of a thrombus is dependent not only on the total concentration of fibrinogen but also on the composition of different isoforms within the fibrinogen pool [5, 6].

3 Fibrinolysis

Understanding fibrinolysis – the process of clot breakdown – is just as essential as understanding coagulation, or clot formation when studying bleeding. The coagulation and fibrinolytic systems are intricately interconnected, working hand in hand to maintain balanced hemostasis. This balance safeguards against both excessive bleeding and pathological clot formation. It is sustained through the collective actions of the fibrinolytic system, natural anticoagulants, and tissue factor pathway inhibitors.

Fibrinolysis ensures the removal of clots once they have accomplished their purpose of sealing vascular injuries. However, if this system is not well-regulated, premature clot dissolution could occur, leading to extended bleeding. In such situations, antifibrinolytic agents like tranexamic acid play a vital role. Large clinical trials, especially in trauma and post-partum hemorrhage, have shown that tranexamic acid effectively prevents severe bleeding by counteracting hyperfibrinolysis.

3.1 Mechanism of Fibrinolysis

Fibrinolysis begins after a blood vessel has healed, involving the breakdown of the clot, or thrombus, facilitated by the enzyme plasmin. Plasmin is derived from its precursor, plasminogen, either on the surface of the fibrin clot or on cellular surfaces. This activation is primarily catalyzed by tissue plasminogen activator (tPA), which is synthesized and released by endothelial cells, and by urokinase plasminogen activator (uPA), produced by monocytes, macrophages, and urinary epithelial cells. Once activated, plasmin digests fibrin into fibrin degradation products (FDPs), some of which have immunomodulatory and chemotactic traits. The activities of tPA and uPA are transient in circulation, lasting only four to eight minutes, as they are swiftly neutralized by specific inhibitors such as plasminogen activator inhibitor-1 (PAI-1) [7].

3.2 Regulation of Fibrinolysis

Fibrinolysis is tightly controlled to ensure that clots are broken down only when necessary. Excessive plasmin activity could lead to indiscriminate clot dissolution, while insufficient activity might result in thrombosis. Several inhibitors, including serine protease inhibitors (serpins), regulate plasmin and plasminogen activators. PAI-1 is the most prominent among these inhibitors, with additional contributions from PAI-2 and α2-antiplasmin, plus others like C1-esterase inhibitors and members of the contact pathway of the coagulation cascade. Another critical regulator is the thrombin-activated fibrinolysis inhibitor (TAFI), activated by thrombin bound to thrombomodulin. Altogether, these mechanisms prevent excessive fibrinolysis and restrict clot breakdown only to areas of vascular healing [7]. This intricate regulation prevents

unnecessary intravascular fibrin accumulation while ensuring thrombi are removed from healed vessels (Figure 3).

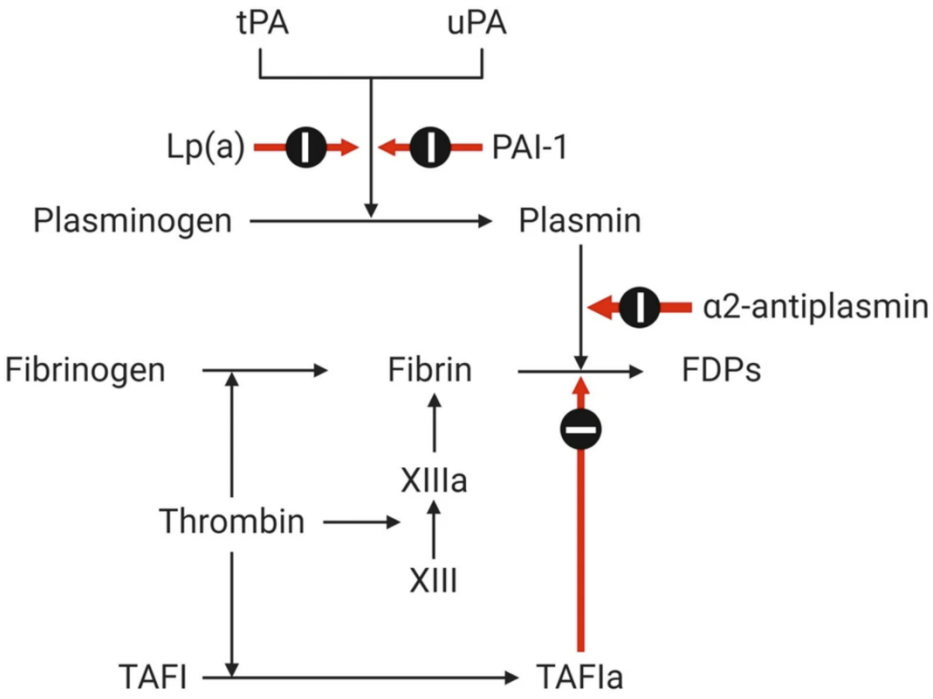

Figure 3 - Effects of different factors on the process of fibrinolysis. FDP – fibrin degradation product, Lp(a) – Lipoprotein A, PAI-1 – plasminogen activator inhibitor 1, TAFI – thrombin activatable fibrinolysis inhibitor, tPA – tissue plasminogen activator, uPA – urokinase plasminogen activator. Adapted from Gue, Y.X., et al., 2021 [8].

The distinctions between fibrinolysis occurring on thrombi and cell surfaces underscore the complexity of these processes. Additionally, fibrinolysis intersects with coagulation at multiple junctures, such as through the inactivation of coagulation factors like FVa and FVIIIa by plasmin. Platelets may also have a role in regulating fibrinolysis, although this remains inadequately comprehended. Moreover, thrombin, upon complexing with thrombomodulin, converts protein C into its anticoagulant form, signifying further interplay between coagulation and fibrinolysis [9].

3.3 Clinical dysregulation of fibrinolysis

Dysregulation of fibrinolysis can manifest either as hyperfibrinolysis or hypofibrinolysis, each of which has notable clinical implications. Hyperfibrinolysis, which is characterized by excessive clot breakdown, is a primary cause of severe bleeding in conditions such as disseminated intravascular coagulation (DIC), trauma-induced coagulopathy, chronic liver disease, metastatic prostate cancer, and certain surgical procedures including cardiovascular procedures [7]. On the

other hand, hypofibrinolysis results in impaired clot dissolution and contributes to thrombotic complications in diseases such as hypothyroidism, multiple myeloma, antiphospholipid syndrome, and alcoholic liver disease [7].

Although congenital fibrinolytic disorders are complex and diverse, they do not constitute the primary focus of this thesis. Instead, the emphasis falls on acquired dysregulations, which are more regularly encountered in clinical practice. These acquired disorders highlight the significance of understanding fibrinolysis's intricacies to steer appropriate therapeutic interventions. Despite substantial advancements in the field, many facets of fibrinolysis continue to be poorly understood. Research is ongoing to address finer aspects of this process, particularly its intersections with coagulation and its role in both bleeding and thrombotic disorders.

4 Fibrinogen: From structure to clinical significance

4.1 Historical overview

For decades, fibrinogen has been recognized as essential for hemostasis, the process that halts bleeding and initiates vessel repair. Its historical journey began in 1666 when Marcello Malpighi, a pioneer in microscopy, discovered fibrin while studying blood clots and cardiac thrombi. Using a light microscope, he observed a fibrous texture within a framework of red blood cells, marking the first identification of fibrin. In 1788, Antoine Fourcroy classified animal matter into three categories: gelatin, albumin, and fibrin, accentuating fibrin's role in blood clotting and its prevalence in muscles. The term "protein" was later introduced by Berzelius in 1838 to describe molecules like fibrin and albumin, derived from the Greek word 'proteíos' to highlight their fundamental role in animal biology.

The precursor of fibrin, fibrinogen, was named in 1847 by Rudolf Virchow, who described it as a "slowly clottable substance" distinct from fibrin. Hermann Schmidt demonstrated in 1872 that the conversion of fibrinogen to fibrin was an enzymatic process, a discovery foundational to our understanding of coagulation. By 1879, Olof Hammarsten had purified fibrinogen for the first time through repeated precipitation, enabling a more in-depth study of its role in hemostasis [1].

These milestones mark the inception of our understanding of fibrinogen. As science has evolved, so too has our appreciation for its multifaceted role in hemostasis, inflammation, and more.

4.2 Structural and functional properties

Fibrinogen, also known as clotting factor 1 (FI), is a 340 kDa glycoprotein that is encoded by three genes (*FGA*, *FGB*, and *FGG*) situated on chromosome 4. This glycoprotein is synthesized in the liver and circulates in the plasma at a concentration of 2–4 g/L, sustaining a half-life of

approximately 4 days. Its structure consists of three sets of non-identical polypeptide chains (α, β, and γ) that are interconnected by disulfide bonds (Figure 4) [5, 6, 10, 11].

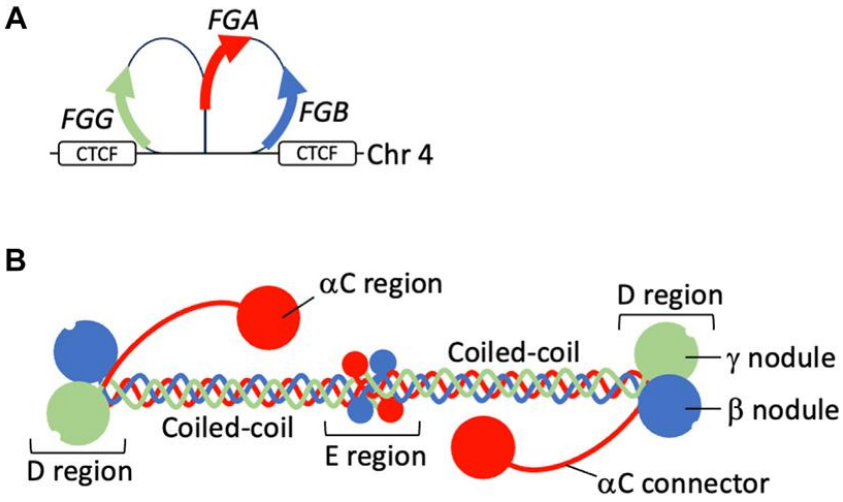

Figure 4 - Structure of Fibrinogen. (A) Fibrinogen is encoded by three genes (*FGA*, *FGB*, and *FGG*), all of which are clustered on human chromosome 4. Fibrinogen is a 340-kDa hexameric glycoprotein that comprises two of each of the following three polypeptide chains: A α (red), Bβ (blue), and γ (green). Adapted from Wolberg A. 2023 [6].

Fibrinogen levels can vary under physiological and pathological conditions. Factors such as age, pregnancy, lifestyle choices (e.g., smoking, exercise), and inflammatory states are associated with increased fibrinogen expression. Conversely, pathological conditions like liver disease and therapeutic interventions, including glucocorticoid administration, may lead to reduced plasma fibrinogen levels [5, 6, 11]. The molecular structure of fibrinogen is crucial to its role in hemostasis. This remarkable glycoprotein is cleaved by thrombin into soluble fibrin monomers, which polymerize to form an intricate fibrin mesh. This network traps red blood cells and platelets, providing the structural basis for clot formation. Fibrinogen also facilitates platelet aggregation by binding to glycoprotein IIb/IIIa receptors on platelet surfaces, thereby linking primary and secondary hemostasis. Beyond its role in coagulation, fibrinogen serves as an acute-phase reactant and modulates inflammatory cellular responses [10, 12].

Variants within fibrinogen structural genes are directly associated with fibrinogen disorders, which are categorized into two main types. Type I deficiencies, such as congenital hypofibrinogenemia and afibrinogenemia, result from a partial or complete absence of circulating fibrinogen. Conversely, Type II mutations cause structural alterations that impair fibrinogen functionality, as seen in dysfibrinogenemia. These mutations may or may not be associated with reduced fibrinogen levels, as observed in hypo-dysfibrinogenemia [5, 6]. Interestingly, genetic polymorphisms within the fibrinogen structural genes only account for about 2% of the variability in plasma fibrinogen concentration. This implies that other factors such

as genes involved in inflammatory pathways or unrelated genetic processes, contribute significantly to fibrinogen variability in the population [5, 6].

Fibrinogen exists in multiple isoforms due to alternative splicing and post-translational modifications. Among these, γ' fibrinogen, an alternative splice variant of the γ chain, represents 8–15% of total fibrinogen in healthy individuals, with plasma concentrations averaging around 0.3 mg/mL. In inflammatory states, particularly those driven by interleukin-6, γ' fibrinogen expression is noticeably upregulated, sometimes along with an increase in total fibrinogen levels. This isoform is associated with the formation of fibrin fibers, which are thinner and present more branching, resulting in a clot that is structurally weaker and more prone to lysis. These alterations in clot architecture might have implications for thrombus stability and its significance in inflammatory diseases [5, 6].

4.3 Fibrinogen in Hemostasis: Challenges and Advances

4.3.1 Surgery and Bleeding

Surgery is often associated with bleeding, which can pose a challenge for hemostasis. During the perioperative period, several factors can lead to bleeding in patients: blood loss, consumption, dilution, acidosis, hypothermia, hyperfibrinolysis, concomitant comorbidities, and medications that may affect hemostasis. These issues can be exacerbated by delayed diagnosis and inappropriate treatment choices. Excessive bleeding is common in patients who undergo cardiac surgery with cardiopulmonary bypass, necessitating the transfusion of allogeneic blood products. This bleeding is influenced by several complex factors including the underlying disease, anticoagulant medication used during the procedure, inflammation, consumption and dilution of clotting factors, and impairment and consumption of platelets associated with cardiopulmonary bypass [13, 14].

Fibrinogen is a coagulation factor that plays a vital role in maintaining both primary and secondary hemostasis. It often becomes the first coagulation factor to reach dangerously low levels in instances of severe bleeding and haemodilution. This situation worsens with hypothermia and acidosis, further highlighting the necessity for its specific replacement [15]. Given its importance and distinctive role in maintaining hemostasis, fibrinogen supplementation becomes critical to manage blood loss control. The body's natural fibrinogen synthesis may be inadequate to compensate for its consumption during acute hemorrhage, where fibrinogen levels are maintained through a delicate balance between synthesis, consumption, and degradation.

Fibrinogen concentrate is frequently used to rectify hypofibrinogenemia during the perioperative period and in other clinical situations, such as trauma and post-partum hemorrhage. Its effectiveness is influenced by several factors, including patient demographics,

comorbidities, associated treatments, the need for other hemostatic agents, the expertise of the surgical team, and the presence of different plasma fibrinogen isoforms that impact clot strength and its susceptibility to fibrinolysis.

While fibrinogen's role in the surgical setting is crucial, its importance extends beyond the operating room. Advances in diagnostics and therapeutic strategies have expanded its use in managing diverse hemorrhagic scenarios, as discussed in the following section.

4.3.2 Fibrinogen in Hemorrhage

The effective management of hemorrhage has seen a paradigm shift in recent decades: moving from broad transfusion strategies to targeted interventions, which are based on individual hemostatic deficits. This evolution has emphasized the central role of fibrinogen in stabilizing clots and controlling blood loss across a variety of clinical contexts.

In bleeding patients, therapeutic decisions now prioritize specific plasma factor concentrates, such as fibrinogen concentrate and prothrombin complex concentrate, over traditional blood component ratios (for example, 1:1:1 transfusions) [16]. This shift is particularly apparent in trauma, obstetrics, and hepatic surgeries, where timely fibrinogen supplementation has proven to improve outcomes. The use of antifibrinolytics, such as tranexamic acid, complements this approach by preventing premature clot dissolution [16, 17].

In many European countries, including Portugal, the use of factor concentrates has increased, particularly fibrinogen concentrate and prothrombin complex concentrate [3]. This trend coincides with a reduction in the use of fresh frozen plasma and other blood components [18].

Point-of-care tests, such as thromboelastography (TEG) and rotational thromboelastometry (ROTEM), have considerably revolutionized the management of hemorrhage. These bedside tests offer immediate insights into the rate of clot development, its stability, and its tendency to dissolve. Initially instituted in 1948 by Hartert, TEG saw a reappearance with contemporary improvements, which led to the creation of ROTEM as a corresponding and commonly utilized diagnostic tool [19, 20]. These technologies are especially helpful in trauma settings, operative rooms, obstetrics, and intensive care units. They count on a small sample of blood (300 µL), to which a consistent rotational force is directed. This method provides a visible and quantitative examination of blood coagulation, documenting clot formation, propagation, stabilization, and eventual dissolution under low-shear conditions similar to those in large veins and the vena cava [19, 21].

Although useful, routine coagulation tests pose several limitations in comparison to point-of-care devices. Notably, they are time-intensive and often lack instantaneous availability. Furthermore, these tests are conducted in plasma, devoid of platelets and other cellular elements crucial in hemostasis. Routine tests also cannot address the impacts of hypothermia and

hyperfibrinolysis, which are significant in bleeding situations. Modern coagulation models merged with real-time diagnostics, have highlighted the fundamental role fibrinogen plays in hemostasis. The focus has now shifted towards avoiding allogeneic blood transfusions by directly addressing the patient's specific hemostatic deficiencies. These advancements underscore fibrinogen as a critical factor in ensuring effective clot formation and stability, especially in conditions influenced by hypothermia and acidosis.

4.3.3 Fibrinogen's role in clinical hemorrhage management

Fibrinogen represents the final step in the coagulation cascade, and its sufficient availability is crucial for forming a stable clot capable of stopping bleeding until vascular wound healing is complete. Over the past decade, the benefits of fibrinogen supplementation have been recognized in various clinical situations, including cardiovascular and vascular surgeries, obstetric complications, trauma, and hepatic surgery [22, 23].

The threshold for fibrinogen administration, measured by the Clauss method, remains a subject of debate. The optimal threshold likely varies across clinical scenarios. Despite this variability, the importance of fibrinogen in modern coagulation pathophysiology is undisputed. Even in cases of thrombocytopenia, fibrinogen plays a crucial role in establishing a robust and functional clot, emphasizing its indispensability in hemostatic management.

The evolving understanding of coagulation mechanisms, in conjunction with advances in diagnostic technologies and therapeutic strategies, has established fibrinogen as a critical component in the management of hemorrhage. Its pivotal role in achieving effective hemostasis, especially in complex clinical scenarios, highlights its significance in contemporary medicine. Through the use of targeted therapies and real-time diagnostics, fibrinogen continues to enhance the outcomes of bleeding patients, marking a substantial improvement in the realm of hemostasis and transfusion medicine.

4.4 Prothrombotic states

Gordon Lowe characterizes thrombosis as "hemostasis in the wrong place", a condition marked by the undue activation of platelets and coagulation factors [10]. Thrombosis is noted in a range of clinical circumstances, with prothrombotic states occasionally developing into full-scale thrombotic episodes.

DIC vividly demonstrates a prothrombotic condition, typically occurring in microcirculation. This condition often emerges as a result of sepsis, leukemia, pancreatitis, liver disease, obstetric complications, or severe tissue injury. DIC leads to fibrinogen consumption, culminating in a delicate balance between thrombosis and bleeding. In opposition, fibrin thrombus formation is more frequently encountered in scenarios such as arterial atherosclerotic plaque rupture,

intracardiac thromboembolism, and venous thromboembolism. These diverse pathophysiological processes highlight fibrinogen's crucial role in coagulation and its implications for thrombotic disease [9].

4.4.1 Fibrinogen and its role in prothrombotic states

Elevated plasma fibrinogen levels have been linked to thrombotic states for a long time and are considered a significant risk factor for thrombotic diseases [9]. Fibrinogen's function as a procoagulant, acute-phase reactant, and agent that increases plasma viscosity and erythrocyte sedimentation rate provides a substantial theoretical foundation for this correlation [10]. In sum, these properties make elevated fibrinogen levels independent predictors of coronary heart disease events. However, despite this strong link, it remains unclear whether fibrinogen or increased viscosity plays a direct causal role in arterial thrombosis.

4.4.2 Inflammation and coagulation activation

Inflammation is another key factor in the activation of coagulation. The mechanisms involved are multifaceted and engage vascular endothelial cells, which play a central role in this process. Cytokines, released by activated leukocytes, promote widespread fibrin deposition within the vasculature, while natural anticoagulants like the protein C and S systems are simultaneously inhibited. Endogenous thrombolysis is impaired by the elevated levels of PAI-1, the primary inhibitor of plasminogen activation. Furthermore, the contact system, typically anticoagulant and profibrinolytic, seems to have a nuanced role in these interactions. These factors collectively highlight the intricate interplay between inflammation and coagulation, with fibrinogen playing a crucial part in this dynamic equilibrium [9, 11].

4.4.3 Epidemiologic insights

Recent epidemiological studies, reviewed by Wolberg and Sang, provide further insights into the role of fibrinogen in thrombosis. Their analysis suggests that elevated fibrinogen levels are positively associated with a higher risk of venous thrombosis among the elderly, an increased risk of cardiovascular diseases such as stroke and coronary heart disease in middle-aged adults, as well as the development of subclinical atherosclerosis in young adults [24]. However, the complexity of these relationships is increased due to numerous confounding factors. Factors such as inflammatory status, interactions with other proteins like Factor XIII, the presence of fibrinogen isoforms (which may weaken the clot), and differences in population demographics, make it difficult to determine the precise contribution of fibrinogen to thrombotic states. While the correlation between fibrinogen and atherothrombogenic is apparent, the exact nature of this connection remains uncertain [5].

4.4.4 Therapeutic considerations of fibrinogen supplementation

In clinical practice, the management of hypofibrinogenemia in bleeding patients is relatively well-established. When bleeding occurs, fibrinogen supplementation is widely seen as a critical intervention for restoring hemostasis. Among the therapeutic options, fibrinogen concentrate is often preferred in urgent or emergencies. It provides a transient but effective increase in plasma fibrinogen levels, which allows for the stabilization of clot formation and the cessation of bleeding [25-28].

However, this approach raises a crucial question: could the administration of exogenous fibrinogen concentrate to correct endogenous hypofibrinogenemia inadvertently create a prothrombotic condition? While some clinical studies have explored this possibility, the persistent influence of confounding variables – such as inflammatory markers, demographic factors, and comorbidities – renders a definitive conclusion elusive.

In summary, fibrinogen supplementation is an indispensable tool in the management of bleeding. However, its potential role in promoting thrombosis remains a significant topic of clinical and research interest. A deeper understanding of these mechanisms is necessary to ensure the safe and effective use of fibrinogen across diverse medical contexts. This knowledge will guide the development of strategies to optimize patient outcomes while minimizing thrombotic risks.

4.5 Management of hypofibrinogenemia

The supplementation of fibrinogen is a crucial intervention in managing bleeding coagulopathies, although it remains a subject of debate. The three primary options for fibrinogen replacement – plasma, cryoprecipitate, and fibrinogen concentrate – each possesses unique advantages and limitations.

4.5.1 Plasma

Although plasma is frequently used, it is not the ideal choice for fibrinogen replacement due to its relatively low fibrinogen concentration. Solvent detergent plasma and fresh frozen plasma (FFP), the most commonly utilized types, generally contain fibrinogen levels between 2 and 2.9 g/L. However, some FFP units might have as low as 1 g/L. Consequently, substantial volumes of plasma are needed to achieve effective fibrinogen correction. In addition, plasma transfusion necessitates ABO compatibility and pre-thawing, a process demanding both time and specialized equipment [25, 26].

4.5.2 Cryoprecipitate

Cryoprecipitate provides a higher fibrinogen content compared to FFP, but it presents challenges related to inconsistency in fibrinogen levels and safety issues. Since it is a pooled product derived from 6 to 10 blood donors, cryoprecipitate increases the recipient's exposure to multiple donors. Additionally, cryoprecipitate does not undergo viral inactivation processes, which raises concerns about its safety. Consequently, it has been withdrawn from regular use in several European countries [29].

4.5.3 Fibrinogen concentrate

Fibrinogen concentrate has emerged as the preferred option for correcting hypofibrinogenemia in many settings. It provides a fixed fibrinogen dose per vial, typically 1 g ranging from 0.9 g to 1.3 g, in a small volume. Furthermore, it undergoes pasteurization and viral inactivation, thus ensuring a high safety profile. Fibrinogen concentrate is rapidly available, eliminates the need for ABO matching, and has not been linked to adverse thromboembolic events in most studies [28, 30]. Dosages for fibrinogen concentrate vary, typically ranging from 25 to 75 mg/kg, depending on the clinical context, viscoelastic testing results, or measurements using the Clauss method [22].

In clinical practice, growing evidence supports the use of fibrinogen concentrate in trauma, post-partum hemorrhage, and perioperative bleeding [31-33]. These studies underscore its role in mitigating acquired coagulopathy and diminishing the necessity for blood transfusion. Both viscoelastic testing and traditional coagulation assays assist clinicians in diagnosing fibrinogen deficits and adjusting interventions. One of the first randomized clinical trials, conducted by Fenger-Erikson, showed that administering 45 mg/kg of fibrinogen to patients undergoing radical cystectomy significantly diminished the need for transfusion of blood products compared to a placebo, regardless of the small sample size [34].

In Portugal, a recent multidisciplinary study has provided updated evidence on managing acquired bleeding coagulopathies in perioperative, obstetric, and trauma settings. This nationwide consensus highlighted fibrinogen concentrate as a preferred therapeutic option for restoring hemostasis in cases of bleeding [35]. The debate surrounding trigger and target fibrinogen levels reflects the complexity of this intervention. Guidelines from bodies such as the European Society of Anaesthesiology (ESA) suggest fibrinogen levels between 1.5 and 2.0 g/L as thresholds for supplementation. However, these targets can vary depending on clinical context and patient-specific factors [36].

The management of acquired coagulopathy should take into consideration the adverse effects of transfusion, including transfusion-related acute lung injury and circulatory overload, both of which are associated with high morbidity and mortality. The dependence on allogeneic

blood products further underscores their scarcity, cost, and associated risks. Consequently, fibrinogen concentrate presents a convincing alternative, minimizing these challenges while efficiently correcting hypofibrinogenemia.

5 Fibrinogen concentrate and safety concerns

While evidence generally supports the efficacy and safety of fibrinogen concentrate, conflicting data persists regarding its optimal use. Variations in trigger values, dosage recommendations, and potential side effects remain unresolved. Most randomized clinical trials and observational studies suggest fibrinogen concentrate is effective and safe, but questions linger about its potential contribution to thrombotic events, particularly in patients with existing risk factors [37-40]. Decades of pharmacovigilance have yielded relatively few adverse event reports, reinforcing its safety profile. Nevertheless, clinicians must remain vigilant, considering the procoagulant nature of fibrinogen concentrate and the cumulative thrombotic risks in coagulopathic patients [41, 42].

The selection of fibrinogen concentrate as a treatment option raises a critical question: why focus on its safety? A distinct factor of human origin, having a well-defined concentration and composition and subjected to viral inactivation, presents an attractive choice for targeted replacement therapy. However, despite its merits and the plethora of published studies, debates about its safety and effectiveness continue to be unresolved.

This work focuses specifically on safety and adverse events, a critical aspect that has not been thoroughly addressed in the existing literature. Adverse events are seldom designated as primary outcomes in clinical studies. Often, they are not pre-specified at all, leading to a lack of clarity in the methods used to gather, define, and report these adverse events.

The varied and inconsistent methods for defining and monitoring adverse events add complexity to this issue. For instance, some trials systematically categorize composite adverse and serious adverse events, while others report multiple categories of adverse events without offering clear definitions. In certain cases, studies only provide the total number of adverse events, with minimal or no detailed data included [43, 44].

Monitoring and reporting adverse events in clinical trials is a complex and nuanced process. It necessitates meticulous blinding of patients and investigators, distinguishing between adverse and serious adverse events, and directly attributing adverse events to study drugs. The collection of accurate patient reports and the maintenance of consistent, transparent monitoring and reporting practices by investigators also heighten the challenge. It is important to acknowledge that adverse events reported during trials may not always be directly linked to the intervention product, even when those events are associated with that drug category. This

consideration is particularly relevant for fibrinogen concentrate, a recognized prothrombotic agent [45].

Given these complexities, this thesis undertakes a targeted analysis of fibrinogen concentrate safety, striving to offer a balanced and comprehensive view of its adverse reactions. Frequently, these adverse reactions are accorded secondary importance in clinical assessments, potentially distorting overall efficacy perceptions. By highlighting the safety profile of fibrinogen concentrate, this work aims to contribute to a more nuanced understanding of its role in clinical practice and facilitate informed decision-making in hemostatic management.

CHAPTER II

Objectives

General objective

The primary objective of this research was to evaluate the safety profile of fibrinogen concentrate, with a focus on its adverse events, to achieve a balanced, evidence-based perspective.

Specific objectives

In detail, the specific objectives were as follows:

1 - Conduct a systematic review with meta-analysis to explore the association between safety outcomes in adult patients – excluding obstetric and trauma cases – who underwent cardiovascular, abdominal, or orthopedic surgery and received fibrinogen replacement during the perioperative phase **(Chapter III)**.

2 - Assess the safety of fibrinogen concentrate when administered within therapeutic ranges during the perioperative period to correct bleeding-associated hypofibrinogenemia in adults (**Chapter IV**).

CHAPTER III

Safety of Fibrinogen Concentrate in Non-Trauma and Non-Obstetric Adult Patients during Perioperative Care: Systematic Review and Meta-Analysis

Journal of
Clinical Medicine

MDPI

Systematic Review

Safety of Fibrinogen Concentrate in Non-Trauma and Non-Obstetric Adult Patients during Perioperative Care: Systematic Review and Meta-Analysis

Manuela Gomes [1], Miguel Ângelo-Dias [2,3], Gonçalo S. Duarte [4,5], Sara Simões Dias [3,6], Sofia Silvério Serra [7] and Jorge Lima [2,3,8,*]

1 Transfusion Medicine Department, Hemovida, Hospital da Luz Lisboa, Luz Saúde, 1500-650 Lisboa, Portugal; amdrgv@gmail.com
2 NOVA Medical School (NMS), Faculdade de Ciências Médicas (FCM), Universidade Nova de Lisboa, 1099-085 Lisboa, Portugal; miguel.dias@nms.unl.pt
3 Comprehensive Health Research Centre (CHRC), NOVA Medical School (NMS), Faculdade de Ciências Médicas (FCM), Universidade Nova de Lisboa, 1099-085 Lisboa, Portugal; sara.dias@nms.unl.pt
4 Clinical Pharmacology, Hospital da Luz Lisboa, Luz Saúde, 1500-650 Lisboa, Portugal; gduarte@edu.ulisboa.pt
5 Laboratory of Clinical Pharmacology and Therapeutics, Faculty of Medicine, University of Lisbon, 1649-004 Lisboa, Portugal
6 ciTechCare—Centre for Innovative Care and Health Technology, Polytechnic of Leiria, 2411-901 Leiria, Portugal
7 Library, NOVA Medical School (NMS), Faculdade de Ciências Médicas (FCM), Universidade Nova de Lisboa, 1099-085 Lisboa, Portugal; sofia.serra@nms.unl.pt
8 Department of Obstetrics and Gynecology, Hospital da Luz Lisboa, Luz Saúde, 1500-650 Lisboa, Portugal
* Correspondence: jorgeramoslima@sapo.pt

Abstract: Background: Low fibrinogen levels are associated with an increased risk of perioperative bleeding. However, there is an ongoing debate over the ideal treatment threshold, the benefits of prophylactic supplementation with fibrinogen concentrate, and the best source of fibrinogen. While fibrinogen concentrate supplementation is being widely used to treat bleeding related to acquired haemostatic deficiencies, there is a lack of evidence regarding its dosage, effectiveness, and safety. This systematic review provides an up-to-date summary of the relationship between fibrinogen concentrate supplementation and safety measures in the perioperative care of non-trauma, non-obstetric adult patients. **Methods**: A comprehensive online search was conducted on PubMed/Medline, EMBASE, Scopus, Web of Science, the Cochrane Database of Systematic Reviews, and the Cochrane Central Register of Controlled Trials. **Results**: This systematic review and meta-analysis encompasses ten studies involving 1391 patients. There was a decreased risk of total thromboembolic events in patients treated with fibrinogen compared to the control (OR 0.65, 95% CI 0.43 to 0.98, I^2 = 0%). In addition, when fibrinogen was used prophylactically, it resulted in shorter ICU stays (MD −1.50, 95% CI −2.64 to −0.36), when set against its therapeutic use. A sensitivity analysis on cardiovascular surgery studies did not reveal any statistically significant difference. **Conclusions**: The use of fibrinogen concentrate in the perioperative care of non-trauma and non-obstetric adult patients may lead to potential benefits.

Keywords: fibrinogen concentrate; perioperative care; hypofibrinogenaemia; meta-analysis; systematic review

Citation: Gomes, M.; Ângelo-Dias, M.; Duarte, G.S.; Dias, S.S.; Serra, S.S.; Lima, J. Safety of Fibrinogen Concentrate in Non-Trauma and Non-Obstetric Adult Patients during Perioperative Care: Systematic Review and Meta-Analysis. *J. Clin. Med.* **2024**, *13*, 3482. https://doi.org/10.3390/jcm13123482

Academic Editor: Pratima Chowdary

Received: 30 April 2024
Revised: 27 May 2024
Accepted: 11 June 2024
Published: 14 June 2024

1. Introduction

Surgery may lead to severe bleeding, which can cause haemorrhagic shock—a potentially fatal condition marked by tissue hypoxia, cellular ischaemia, and eventual organ dysfunction, potentially leading to organ failure and death [1]. Fibrinogen, also known

as clotting factor I, is a liver-produced glycoprotein vital to both primary and secondary haemostasis. When thrombin cleaves it, fibrinogen produces soluble fibrin monomers, which polymerise to create a clot base and promote platelet aggregation through binding to glycoprotein IIb/IIIa receptors on platelets. Consequently, fibrinogen proves critical for effective blood clotting and is the first factor drained in cases of extreme bleeding and haemodilution, a situation worsened by hypothermia and acidosis [2]. Researchers link declining fibrinogen levels during bleeding to increased morbidity and mortality rates across various clinical situations, including trauma-induced coagulopathies [3–5]. Unsurprisingly, given fibrinogen's distinct role in sustaining haemostasis, its administration has proven consistently effective in animal models and patients with acquired fibrinogen deficiency, becoming a critical step to control blood loss in critical settings [6]. In these situations, standard coagulation tests, or, preferably, viscoelastic testing, can corroborate clinical suspicions of a fibrinogen deficit. Normal plasma levels extend from 1.5 to 4.5 $g \cdot L^{-1}$ but can naturally be higher [7,8].

Fibrinogen concentrate (FC) is a widely used haemostatic agent in many countries, with several decades of pharmacovigilance yielding relatively few reports of adverse events [9], but fibrinogen replacement is still a matter of debate for several reasons. Firstly, when it comes to supplementation, there are three options: plasma, cryoprecipitate, and FC. Plasma is not the best source for fibrinogen replacement, whereas cryoprecipitate has variable fibrinogen levels and contains other coagulation factors. FC, however, is advantageous since it has a fixed amount of fibrinogen in a small volume (1 g per vial, potentially ranging from 0.9 g to 1.3 g), undergoes pasteurisation and viral inactivation, and is readily available [10,11]. However, FC may not be available in all countries for treating acquired coagulopathies. The suggested doses to correct hypofibrinogenaemia in bleeding patients with coagulation disorders range from 25 to 75 $mg \cdot kg^{-1}$, dependent upon various factors, including the outcome of viscoelastic testing or the Clauss method [12]. Secondly, selection and target levels bring another layer of discussion and appear to rely on different factors and clinical circumstances [13]. Lastly, FC is a procoagulant given to patients with coagulation disorders who already have accumulated risk factors for developing thrombotic or thromboembolic episodes. Although numerous clinical trials suggest that FC is effective and seemingly safe, reliability is curtailed by the vast heterogeneity and low quality of the studies conducted, and their small sample sizes, limiting the rigorous recommendation of FC use in minimising blood transfusion requirements [14–16].

Several published review studies have offered an insight into the impact of FC on bleeding patients, but there are noteworthy limitations. Firstly, meta-analyses by Fominskiy and Wikkelso [14,16] include both paediatric and adult populations, which could skew findings, as fibrinogen levels are known to vary with age [17]. Secondly, Fominskiy et al.'s [16] inclusion of a post-partum haemorrhage study could add heterogeneity, due to pregnancy-related alterations in fibrinogen levels. Both these meta-analyses also include trauma patients whose bleeding is influenced by multiple factors such as tissue injury and shock, thereby potentially skewing mortality outcomes and affecting the reliability of conclusions [5,18]. In the third place, despite Ka Ting et al.'s meta-analysis [15] intending to minimise such heterogeneity, its search databases may not have been comprehensive enough, possibly resulting in overlooked pertinent studies. Lastly, other systematic reviews with meta-analyses include limited studies or only focus on specific clinical settings, surgeries, and comparators, thus lacking a comprehensive evaluation of FC effectiveness and safety in bleeding patients [19–21].

Future studies need to meticulously gather and analyse all existing data on FC in patients with acquired hypofibrinogenaemia who bleed. It is necessary to thoughtfully choose suitable populations and comparators and consider the flaws of prior studies to obtain more precise and dependable conclusions. This systematic review primarily aimed to explore the association between safety outcomes in adult patients—excluding obstetric and trauma cases—who underwent cardiovascular, abdominal, or orthopaedic surgery and received fibrinogen replacement during the perioperative phase.

2. Materials and Methods

This study was preregistered in PROSPERO (No: CRD42023395383) and conducted following the guidelines of the Preferred Reporting Items for Systematic Review and Meta-Analysis (PRISMA) statement [22,23] (online Supporting Information Table S1).

2.1. Information Sources and Search Strategy

On 3 March 2023, a comprehensive computerised literature search was conducted across various electronic databases, namely PubMed/Medline, EMBASE, Scopus, Web of Science, the Cochrane Database of Systematic Reviews, and CENTRAL. We used a search strategy involving database-specific subject heading terms along with their free-text word variants, accounting for the unique features of each database (online Supporting Information Table S2). These were then combined using the Boolean operators 'OR' and 'AND'. The search was limited to studies involving human adults; there were no restrictions on the publication date or language. In addition, a manual review of the references from the chosen studies was carried out to find any pertinent publications that might have been missed. The search was updated in December 2023, and no additional eligible articles were found.

Rayyan Intelligent Systematic Review software was utilised to store, organise, and manage all references resulting from the literature search [24].

2.2. Eligibility Criteria

The inclusion criteria were as follows: the study must be an RCT; the population must consist of patients undergoing surgical care in cardiovascular, abdominal, or orthopaedic surgeries who are experiencing bleeding during the perioperative period; the intervention must involve FC supplementation in comparison to standard treatments, placebos, or other haemostatic agents; and the outcomes must evaluate the safety of FC supplementation.

The exclusion criteria were as follows: studies involving patients with hereditary bleeding disorders leading to fibrinogen deficiency or abnormal function were excluded, as were studies focusing on trauma or obstetric patients, as these were deemed incorrect populations. Studies presented only in abstract form without accompanying full-text publications, individual case studies, editorials, commentaries, letters to the editor, and review articles were not included, as these were considered the wrong publication type. Duplicate studies were also excluded.

2.3. Study Selection

Two authors (MG, MAD) independently screened the titles and abstracts, with any conflicts resolved by a third reviewer (JL). The remaining studies were full text examined by two authors (MG, MAD), and selection was based on the eligibility criteria. In case of disagreements, a third reviewer (JL) was consulted to achieve consensus. All decisions, as well as reasons for exclusion, were recorded. The number of articles chosen at each stage is depicted in a flow chart in compliance with the PRISMA 2020 guidelines. The review authors had access to information about journal titles, study authors, and their institutions.

2.4. Data Collection

Two independent reviewers (MG, MAD) extracted all pertinent data from each chosen study. Only relevant information from studies assessing multiple outcomes and variables was collected for this review. For missing or unreported data, the respective authors of the included studies were contacted for clarification. Any disagreements were resolved by consulting a third reviewer (JL). Using an extraction form, two authors (MG, MAD) independently extracted all relevant data from each study, namely, the characteristics of the study (title, authors, year of publication, journal, country of origin, study design, number of participants and their characteristics, and recruitment procedure and duration), characteristics of intervention (indication, timing, type of surgery, criteria for FC administration, dose of FC administered, and type and dose of comparator), and outcomes.

Data were recorded using Microsoft Excel™ [25].

2.5. Outcomes

The primary outcome was overall mortality. To analyse this outcome, we used the data from the longest follow-up period reported for mortality in each study, regardless of the specific duration across the studies. This period did not exceed 30-day mortality in any of the studies. Secondary outcomes were composite adverse and serious adverse events, and specific adverse events associated with fibrinogen concentrate intervention, namely, thromboembolic events, myocardial infarction, deep venous thrombosis or pulmonary embolism, stroke or transient ischaemic attack, the total length of stay in hospital, and the length of stay in the ICU. Adverse events and serious adverse events were defined by each study's authors and observed and assessed by safety outcome assessors in the operating theatre or postanaesthetic care unit, or during the follow-up period after surgery, depending on each study. The risk of each outcome was measured as the number of patients experiencing a given outcome. This review followed a confirmatory approach, as pre-specified in the protocol, in which anticipated or already recognised specific adverse events associated with the intervention and assumed to be measured regularly and consistently in studies were chosen [26].

Only studies reporting outcome data of interest were considered eligible. We did not extract data as 'zero' unless they were clearly listed as such in the report. No interpretation of withdrawals as surrogate markers for safety or tolerability was adopted [26].

2.6. Risk of Bias

The Cochrane risk of bias tool was utilised to evaluate the methodological quality of each study independently [27,28]. A second reviewer (GSD) examined this evaluation, with any disagreements settled by a third independent reviewer (MG). This tool categorises studies into a low, high, or unclear risk of bias in areas such as random sequence generation, allocation concealment, blinding of participants and personnel, blinding of outcome assessment, incomplete outcome data, and selective reporting [27,28]. We also checked for industry sponsorship or conflicts of interest.

2.7. Statistical Analysis

We utilised R statistical software (version 4.3.0) and package metafor [29] for our statistical analysis and the creation of forest plots. The pooling of our data was executed using a random effects model, given the anticipated heterogeneity in the included trials, mainly the differences in study design. We used an empirical Bayes estimator to pool data. We reported pooled dichotomous data through odds ratios (ORs) and mean difference (MD) for continuous data, with 95% confidence intervals (95% CIs) for both. When trials presented continuous data as the median with IQR, the values were converted to the mean and SD, following the Cochrane methodology [30]. Heterogeneity was examined using tau^2 and I^2 [31]. Subgroup analyses were conducted, considering the comparator group (cryoprecipitate, platelets, placebo, or no treatment) and whether the intervention was prophylactic or therapeutic. Post hoc sensitivity analyses were performed for all outcomes by including only data from cardiovascular surgery trials.

For the analysis of publication bias, we conducted a linear regression of funnel plot asymmetry using Egger's test [32]. Statistical significance was considered at $p < 0.05$.

3. Results

3.1. Study Selection

The database search yielded 2542 studies, from which 653 were dismissed as duplicates. We found an additional seven studies by a manual search of references lists. After screening the remaining 1882 records by title and abstract, we excluded 1818 studies that evidently did not meet our eligibility criteria. We identified 64 potentially relevant records for a full review. Eventually, we acquired and examined 59 full-text articles, only to exclude

another 49 due to inapplicable publication type (26 records), non-RCT studies (15 records), irrelevant outcomes (5 records), or a not-of-interest study population (3 records).

Ten studies fulfilled all criteria and were selected for systematic review and meta-analysis [33–42]. Figure 1 illustrates the process of study selection.

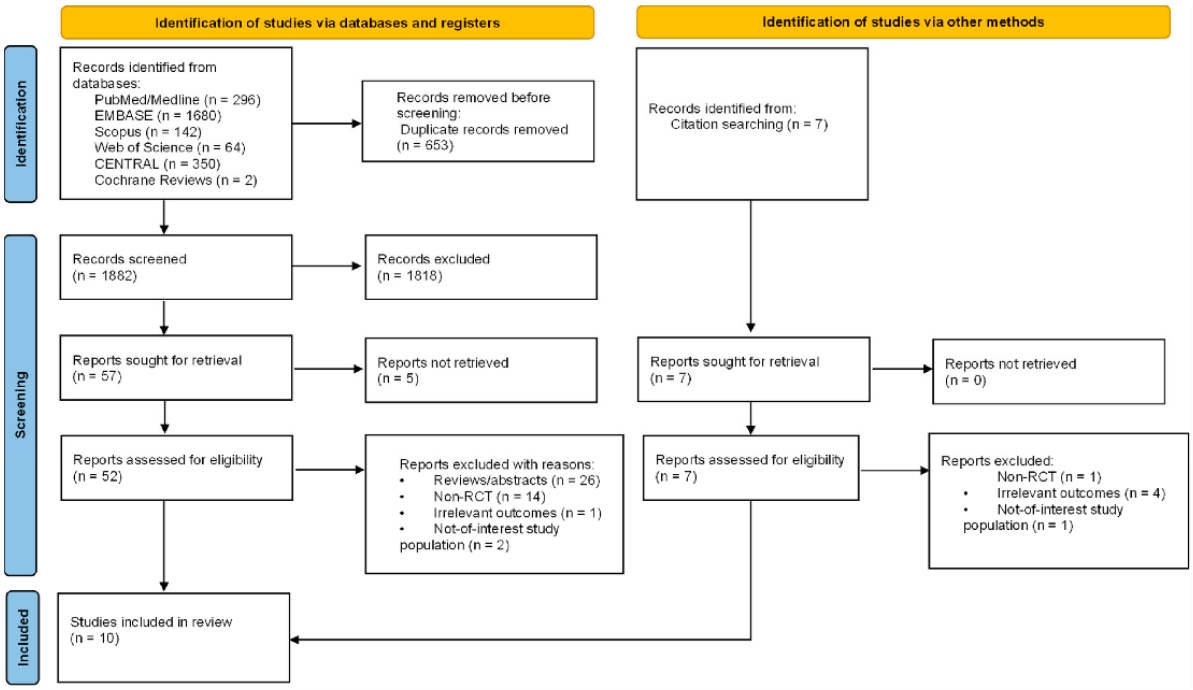

Figure 1. PRISMA 2020 flow diagram of systematic review process and study selection.

3.2. Characteristics of Included Studies

Table 1 summarises the features of the included studies. These articles, published from 2009 to 2019, originate from 16 different countries.

Among the studies, seven focused on cardiovascular surgery [33–35,39–42], one on elective liver transplantation [37], one on cytoreductive surgery [36], and one on hip arthroplasties [38]. The goal of evaluating FC administration in treating bleeding patients varied across the trials. Four studies examined the advantages of administering FC as a first-line treatment pre-operation or during surgery, to prevent bleeding and reduce postoperative blood loss and transfusion needs [37–40]. Three studies evaluated the benefits of using a single intra-operative therapeutic bolus of FC to control bleeding after complicated cardiac surgery, without selection based on plasma FC level [34,35,42]. Three studies aimed to juxtapose therapeutic FC administration with other haemostatic agents to treat acquired hypofibrinogenaemia [33,36] or poor haemostatic conditions like moderate to severe bleeding [41].

The criteria for FC administration varied across the trials reviewed. Three studies used specific plasma fibrinogen levels as criteria: Karlsson [39] set the limit at pre-operative fibrinogen $\leq$3.8 g·L^{-1}, Sabate [37] used pre-operative fibrinogen $\leq$ 2.9 g·L^{-1} or intra-operative levels < 1 g·L^{-1}, while Callum [33] considered intra-operative fibrinogen < 2.0 g·L^{-1} or the clot amplitude at 10 min < 10 mm by thromboelastometry as criteria. Three other studies applied a bleeding mass criterion, specifically 60–250 g [34,35] or 60–250 mL [42] within the first 5 min after removal from cardiopulmonary bypass (CPB). One trial administered FC according to predicted blood loss during surgery; FC was administered when the blood loss was projected to surpass 2 L when 90 min into the procedure [36]. Another study started FC administration when the visible bleeding scale reached 2 (controllable bleeding with applied pressure) or 3 (multiple diffuse bleeding sites) [41]. Two remaining studies did not establish any predefined criteria for FC administration [38,40].

Table 1. Characteristics of included studies.

Study	Country	Trial Duration	Intervention Group	Indication	Timing	Type of Surgery	Criteria for FC Administration	Administered FC Dose	Comparator Group	Comparator
Bilecen et al., 2017 [42]	The Netherlands	Feb 2011–Jan 2015	n = 60 70.0 (10.0) yrs 65% males	Therapeutic	Intra- or post-op	Elective high-risk cardiac surgery *	5-min bleeding mass of 60–250 mL immediately after CPB and completion of surgical haemostasis	Fg doses based on plasma Fg levels (Clauss) at end of CPB †. (Haemocomplettan-P®, CSL Behring)	n = 60 72.0 (10.0) yrs 73% males	Placebo (2 g of albumin diluted with 0.9% saline solution)
Callum et al., 2019 [33]	Canada	Feb 2017–Nov 2018	n = 372 65 (54–72) yrs 69.6% males	Therapeutic	Intra or post-op	Cardiac with CPB	Plasma Fg < 2.0 $g \cdot L^{-1}$ or FIBTEM–A10 < 10 mm	4 g FC (Fibryga®, Octapharma AG)	n = 363 64 (53–72) yrs 71.1% males	10 units of cryoprecipitate (~4 g Fg)
Karlsson et al., 2009 [39]	Sweden	Sep–Dec 2006	n = 10 66 (9) yrs 90% males	Prophylactic	Pre-op	Elective coronary artery bypass graft	Plasma Fg ≤ 3.8 $g \cdot L^{-1}$	2 g FC (Haemocompletan®, CSL Behring)	n = 10 68 (8) 90% males	No treatment
Najafi et al., 2014 [38]	Iran	Aug 2011–Feb 2013	n = 15 52.8 (14.9) yrs 46% males	Prophylactic	Intra-op	Total hip arthroplasty	Prophylactic administration	30 mg/kg FC (Haemocomplettan®, CSL Behring)	n = 15 51.3 (14.1) yrs 20% males	Placebo (0.9% saline solution)
Rahe-Meyer et al., 2016 [35]	Austria, Brazil, Canada, Czech Republic, Denmark, Finland, Germany, Italy, Japan, Poland, UK	Jan 2012–Sep 2014	n = 78 63.9 (22–86) yrs 76.9% males	Therapeutic	Intra-op	Elective aortic surgery requiring CPB with or without other cardiac surgery	5-min bleeding mass of 60–250 g immediately after CPB and completion of surgical haemostasis	Fg doses determined from MCF of FIBTEM ‡ (Haemocomplettan-P® and RiaSTAP™; CSL Behring)	n = 74 64.2 (24–86) yrs 68.9% males	Placebo (0.9% saline solution)
Rahe-Meyer et al., 2013 [34]	Germany	Jun 2008–Apr 2010	n = 29 59 (14) yrs 66% males	Therapeutic	Intra-op	Elective aortic replacement surgery involving CPB	5-min bleeding mass of 60–250 g immediately after CPB and completion of surgical haemostasis	Fg doses determined from MCF of FIBTEM ‡ (Haemocomplettan-P® and RiaSTAP™; CSL Behring)	n = 32 61 (12) yrs 78% males	Placebo (0.9% saline solution)
Ranucci et al., 2015 [40]	Italy	Nov 2011–2014	n = 58 72 (64–76) yrs 71% males	Prophylactic	Intra-op	Complex cardiac surgery with expected CPB duration ≥ 90 min	Prophylactic administration	FC doses determined on MCF at FIBTEM ‡ (Haemocomplettan®, CSL Behring)	n = 58 73 (68–79) yrs 74% males	Placebo (0.9% saline solution)

Table 1. *Cont.*

Study	Country	Trial Duration	Intervention Group	Indication	Timing	Type of Surgery	Criteria for FC Administration	Administered FC Dose	Comparator Group	Comparator
Roy et al., 2019 [36]	UK	Mar 2017–Jul 2018	n = 22 61 (37–76) yrs 40.9% males	Therapeutic	Intra- or post-op	Cytoreductive surgery for Pseudomyxoma peritonei	Predicted intraoperative blood loss ≥ 2 L 90 min into the surgery	4 g FC (Fibryga®, Octapharma AG). Intra- or post-op FIBTEM A20 of ≤12 mm triggered administration of another 4 g/2 g of FC	n = 23 59 (34–72) yrs 47.8% males	Cryoprecipitate (2 × 5 units, ~4.3 Fg). Intra- or post-op FIBTEM A20 of ≤12 mm triggered administration of more 10/5 units
Sabate et al., 2016 [37]	Spain	Aug 2012–Feb 2014	n = 48 54.5 (49–60) yrs 81.3% males	Prophylactic or therapeutic	Pre, intra, or post-op	Elective liver transplantation surgery	Pre-op plasma Fg level ≤ 2.9 g·L^{-1}, or intra-op Fg levels < 1 g·L^{-1}	1 g Fg until reaching plasma value of 2.9 g·L^{-1}. (CSL Behring)	n = 44 57 (50–64) yrs 75% males	Placebo (0.9% saline solution)
Tanaka et al., 2013 [41]	USA	Jan 2011–May 2013	n = 10 71.3 (5.3) yrs 80% males	Therapeutic	Intra-op	Elective valve cardiac surgery with CPB	Visual bleeding scale of 2 or 3 §	4 g of Fg (RiaSTAP®, CSL Behring)	n = 10 66.1 (8.9) yrs 80% males	1 unit of apheresis PLTs

Data presented as mean (SD), mean (range), or median [IQR]. FC, fibrinogen concentrate; PLT, platelet; Fg, fibrinogen; MCF, maximum clot firmness of the fibrin-based clot; FIBTEM, fibrin-based thromboelastometry test; CPB, cardiopulmonary bypass. * Defined as combined coronary artery bypass graft (CABG) surgery and valve repair or replacement surgery, the replacement of multiple valves, aortic root reconstruction, or reconstruction of ascending aorta or aortic arch. † Fibrinogen concentrate dose (g) = (2.5 − [Plasma Fibrinogen Level at End of CPB, g/L]) × 0.07 × (1 − Hematocrit on CPB) × BodyWeight (kg). The target was 2.5 g·L^{-1}. ‡ Fibrinogen concentrate dose (g) = (target FIBTEM MCF − actual FIBTEM MCF [mm]) − (body weight [kg]/70 × 0.5 g/mm). The target FIBTEM MCF was 22 mm. § Visual bleeding scale: 0 = excellent haemostasis (dry field), 1 = mild bleeding (oozing), 2 = moderate bleeding (controllable with applied pressure), and 3 = severe bleeding (multiple diffuse bleeding sites).

Three studies utilised set doses of FC at 2 g and 4 g [33,39,41], one study used a fibrinogen dose determined by body weight (30 mg fibrinogen·kg^{-1}) [38], and another determined by plasma fibrinogen levels at the end of CPB and body weight, and corrected for haematocrit [42]. Three additional studies figured the fibrinogen dose based on the ROTEM/FIBTEM maximum clot firmness parameter [34,35,40]. One study administered 1 g of fibrinogen, aiming to reach a plasma value of 2.9 g·L^{-1} [37], and a different study began with a 4 g dose of FC. If the FIBTEM A20 parameter remained ≤12 mm, they supplemented it with a further 4 g dose [36].

Two studies utilised Octapharma's Fibryga® [33,36], while eight others employed CSL Behring's Haemocomplettan®/RiaSTAP® [34,35,37–42] as FC products. Six studies compared the results with a placebo, either isotonic saline [34,35,37,38,40] or saline solution with albumin [42]; two used cryoprecipitate for comparison [33,36]; one compared with apheresis platelets [41]; and one did not provide any treatment for comparison [39].

The study included 702 patients in the intervention group and 689 in the comparison group. Sample sizes ranged from 10 to 372 patients.

3.3. Risk of Bias

Table 2 displays the risk of bias assessment results of the included studies.

Table 2. Methodological quality assessment according to Cochrane risk of bias tool.

Items	Random Sequence Generation	Allocation Concealment	Performance	Detection	Attrition	Selective Reporting	Overall
Bilecen et al., 2017 [42]	●	●	●	●	●	●	●
Callum et al., 2019 [33]	●	●	●	●	●	●	●
Karlsson et al., 2009 [39]	●	●	●	●	●	●	●
Najafi et al., 2014 [38]	●	●	●	●	●	●	●
Rahe-Meyer et al., 2013 [34]	●	●	●	●	●	●	●
Rahe-Meyer et al., 2016 [35]	●	●	●	●	●	●	●
Ranucci et al., 2015 [40]	●	●	●	●	●	●	●
Roy et al., 2019 [36]	●	●	●	●	●	●	●
Sabate et al., 2016 [37]	●	●	●	●	●	●	●
Tanaka et al., 2013 [41]	●	●	●	●	●	●	●

● Green: low risk of bias; ● Red: high risk of bias; ● Yellow: unclear risk of bias.

Seven studies used a computer-generated method for random sequence allocation [33–35,37,38,40,42], while three did not disclose the method used [36,39,41]. Almost every selected study adequately concealed allocation (usually with unmarked, sealed opaque envelopes or syringes), with one not giving this information [38].

The blinding practices used varied across the studies we reviewed. Five studies ensured that participants, caregivers, and outcome assessors were adequately blinded [34,35,37,40,42]. In one study, the clinicians giving the treatment were not blinded; however, participants, data collectors, and outcome assessors were blinded [33]. Two studies did not specifically mention if the participants were blinded, but all other involved staff and outcome assessors were [37,39]. Another study acknowledged that clinician blinding was impractical during surgeries due to noticeable differences in study drugs [36]. One study was conducted as open-label [41] and another as single-blind [38].

Four studies addressed intention-to-treat analyses and covered methods for handling missing data [34,35,37,42]. One study reported that participants were lost during follow-up or removed after randomisation, but the missing data were proportionate across groups, and the reasons were unrelated to the outcome [33]. Another study removed two partici-

pants (4.4%), each from a different group, due to significant protocol deviations (the FC group was underdosed, and the comparison group was overdosed) [36]. In four studies, no participants were excluded following randomisation [38–41].

Concerning selective reporting bias, one study failed to report an outcome specified in their protocol—namely, the volume of transfused packed red blood cells during and post-surgery [38]. Another study did not provide a trial registration or protocol number in its manuscript [39]. There were no suspicions of selective reporting in the other studies, as each outcome reported adhered to the established protocols.

Nine trials were sponsored by the respective FC supplier. One study did not provide such information [38]. Five studies disclosed conflicts of interest [33–36,40], three studies stated no conflict of interest [37,41,42], and two studies did not provide such information [38,39].

Overall, three out of the ten trials included were deemed to have a high risk of bias. This was due to the lack of assurance that the participants and personnel in these three trials were blinded to the received intervention [36,38,41].

3.4. Thromboembolic Events

Data from ten trials (n = 1391) were included in this analysis. The combined analysis revealed that patients treated with fibrinogen had a reduced risk of total thromboembolic events compared to the control (OR 0.65, 95% CI 0.43 to 0.98, I^2 = 0%, Figure 2). A subgroup analysis considering the control group did not yield statistically significant results (interaction *p*-value = 0.88).

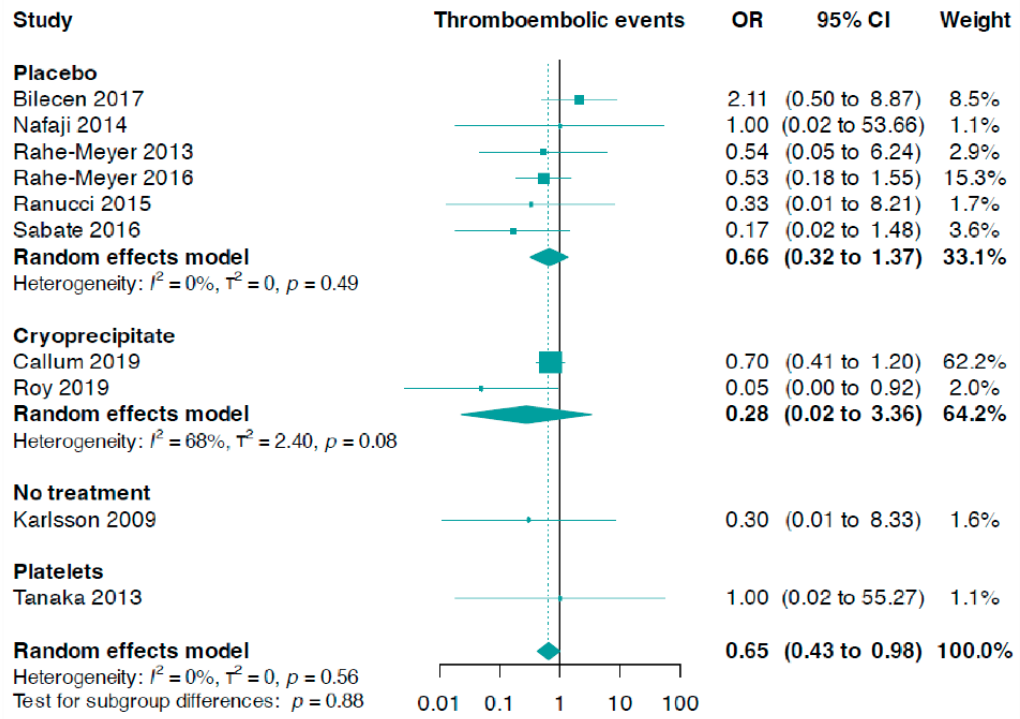

Figure 2. Forest plots for thromboembolic events according to comparator group [33–42].

Additionally, we found no statistically significant difference between fibrinogen and controls in stroke or transient ischaemic attack (OR 0.91, 95% CI 0.51 to 1.60, I^2 = 0%, six trials, n = 1214, online Supporting Information Figure S1). This also applied to myocardial infarction (OR 0.98, 95% CI 0.37 to 2.61, I^2 = 0%, seven trials, n = 1102, online Supporting Information Figure S2), as well as deep venous thrombosis or pulmonary embolism (OR 0.55, 95% CI 0.24 to 1.26, I^2 = 0%, nine trials, n = 1371, online Supporting Information Figure S3).

The subgroup analysis, which focused on fibrinogen administration indications, did not produce statistically significant results (p-value for interaction = 0.26, Figure 3). We discovered that prophylactic fibrinogen use (OR 0.28, 95% CI 0.06 to 1.21, I^2 = 0%) was not different from its therapeutic use (OR 0.69, 95% CI 0.37 to 1.30, I^2 = 13%) in diminishing the risk of thromboembolic events (Figure 3). In addition, there were no notable differences concerning the risk of stroke or transient ischaemic attack (p-value for interaction = 0.94, online Supporting Information Figure S4), myocardial infarction (p-value for interaction = 0.62, online Supporting Information Figure S5), and deep venous thrombosis or pulmonary embolism events (p-value for interaction = 0.60, online Supporting Information Figure S6).

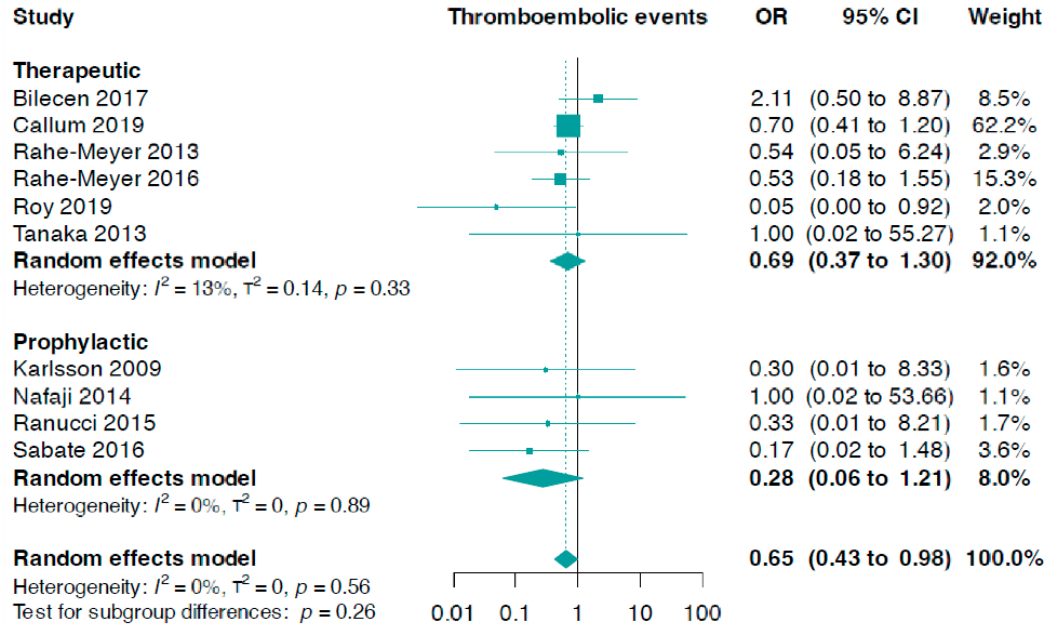

Figure 3. Forest plots for thromboembolic events according to indication setting [33–42].

We also conducted a linear regression of funnel plot asymmetry using Egger's test, which did not indicate evidence of publication bias for thromboembolic events (p-value = 0.125).

3.5. Mortality

Data for this outcome were contributed by ten trials (n = 1391). The pooled analysis indicated there was no meaningful statistical difference between fibrinogen and controls (OR 0.98, 95% CI 0.62 to 1.55, I^2 = 0%, Figure 4). However, no significant statistical difference was observed from a subgroup analysis based on the control group (p-value for interaction = 0.26). Nevertheless, fibrinogen usage showed a slightly, but not statistically significant, lower mortality in comparison to a placebo (OR 0.39, 95% CI 0.14 to 1.07, I^2 = 0%). In addition, there were no significant statistical differences in comparisons of fibrinogen versus cryoprecipitate, fibrinogen versus platelets, or fibrinogen versus no treatment (Figure 4).

The subgroup analysis, based on the need for fibrinogen administration, was not statistically significant (interaction p-value = 0.36, online Supporting Information Figure S7).

We also conducted a linear regression of funnel plot asymmetry using Egger's test, which did not indicate evidence of publication bias for overall mortality (p-value = 0.074).

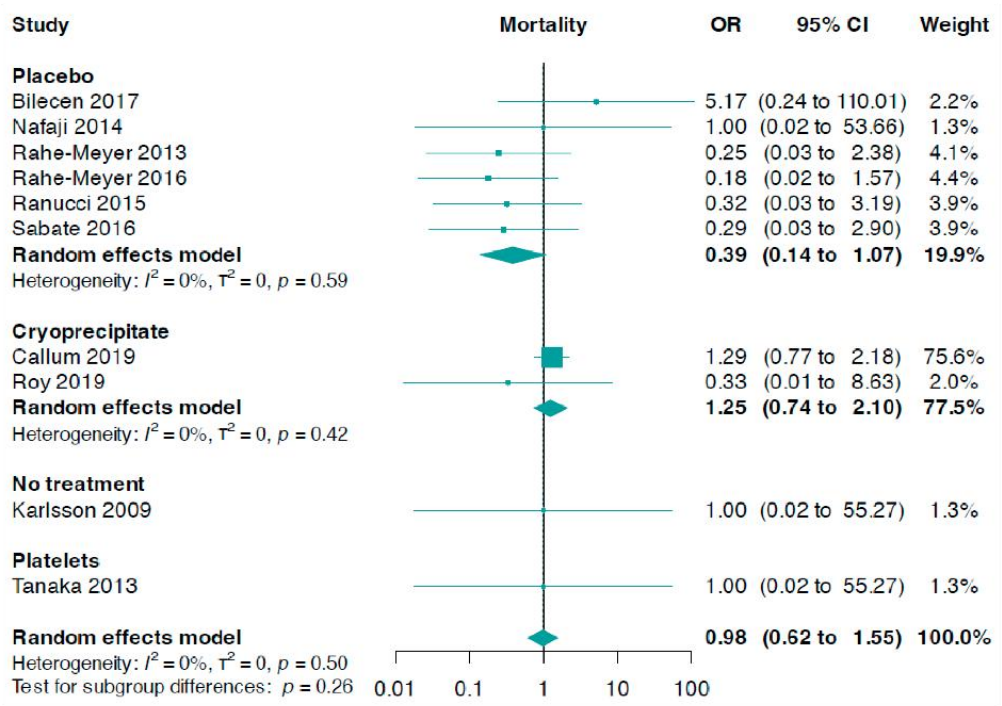

Figure 4. Forest plots for mortality according to comparator group [33–42].

3.6. Length of Hospital Stay

The combined analysis did not reveal a meaningful statistical discrepancy in hospital stays between fibrinogen and controls (MD −0.61, 95% CI −1.36 to 0.14, I^2 = 0%, five trials, n = 839, online Supporting Information Figure S8), nor in ICU duration (MD −0.48, 95% CI −1.13 to 0.18, I^2 = 48%, five trials, n = 912, online Supporting Information Figure S9).

A subgroup analysis based on the reasons for fibrinogen administration revealed significant results (interaction p-value = 0.02, Figure 5). The data showed that the prophylactic use of fibrinogen resulted in a reduction in ICU stay duration (MD −1.50, 95% CI −2.64 to −0.36) compared to the therapeutic use of fibrinogen (MD −0.03, 95% CI −0.46 to 0.41, I^2 = 0%, Figure 5).

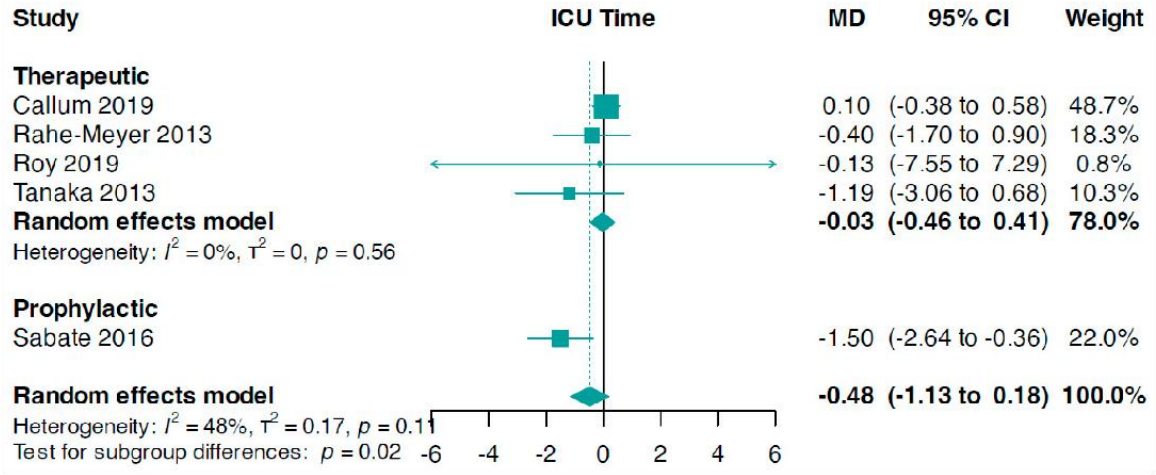

Figure 5. Forest plots for ICU time according to indication setting. MD, mean difference [33,34,36,37,41].

3.7. Safety Outcomes

Data for this outcome were contributed by nine trials (n = 1371). The combined analysis revealed a slightly, but not statistically significant, reduced risk of composite

adverse events in patients treated with fibrinogen compared to the control group (OR 0.79, 95% CI 0.61 to 1.02, I^2 = 0%, Figure 6). In addition, there was no significant statistical difference between the fibrinogen group and the control group concerning serious adverse events (OR 0.65, 95% CI 0.37 to 1.16, I^2 = 53%, five trials, n = 1085, online Supporting Information Figure S10).

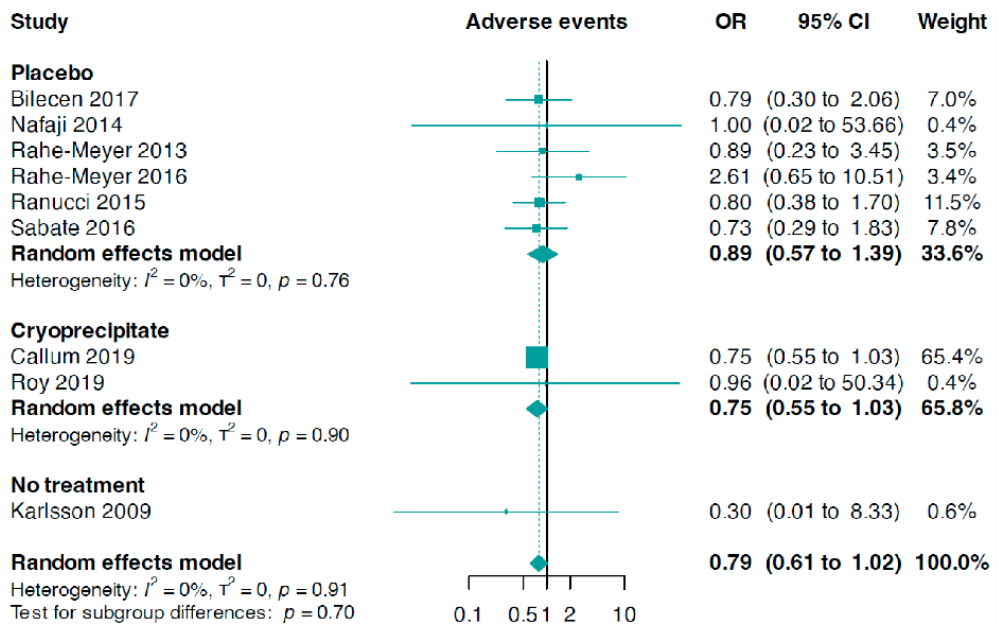

Figure 6. Forest plots for adverse events according to comparator group [33–40,42].

The subgroup analysis, depending on the control group or the reason for administering fibrinogen (online Supporting Information Figures S11 and S12), did not yield statistically significant results.

3.8. Sensitivity Analysis on Cardiovascular Studies

Since most of the included trials were on cardiovascular surgery (seven trials, n = 1224), we performed a sensitivity analysis for all outcomes focusing exclusively on these trials. However, we found no statistically significant differences for any of the outcomes investigated (online Supporting Information Table S3)

4. Discussion

Excessive blood loss during and after surgery is a common, critical complication. It can lead to extended stays in the ICU, increased reliance on mechanical ventilation, heightened health risks and death rates, and elevated healthcare costs [43,44]. Fibrinogen, a vital component for achieving and maintaining haemostasis, significantly decreases during bleeding [6]. As a result, FC is increasingly used worldwide to control bleeding in patients with acquired hypofibrinogenaemia. However, robust evidence supporting its safety is still limited.

Our systematic review of ten studies involving 1391 patients highlights a decreased overall risk of thromboembolic events after administering FC in perioperative settings, and a reduction in ICU stay when used prophylactically. Additionally, although our analysis showed a small and potentially beneficial effect of FC intervention on overall mortality, particularly when compared to a placebo, and a reduced risk of composite adverse events, these observations were not statistically significant. Thus, more trials are needed to substantiate these findings. Nevertheless, these observations are crucial for haemostatic agents in light of past cases where products like Factor VIIa effectively minimised blood loss but raised safety issues due to thrombosis and mortality risks [45,46].

To our understanding, this study offers the most thorough and current meta-analysis, focusing on the safety of FC use in specific non-trauma, non-obstetric adult patients during perioperative care.

In this study, we chose to analyse the risk of adverse events, to achieve a balanced perspective, as these often take second place in the evaluation of interventions and may make the intervention look more favourable than it should. However, the monitoring and reporting of adverse events in a clinical trial is a complex task, involving various considerations. This includes ensuring the proper blinding of both patients and investigators, distinguishing between adverse and serious adverse events, attributing adverse events to study drugs, collecting accurate reports from patients, and maintaining consistent, transparent monitoring, coding, and reporting by investigators. In addition, some adverse effects occur rarely or may become apparent long after the start of the intervention. This contrasts with adverse events that have a higher incidence and occur soon after the intervention is delivered. Therefore, it is important to recognise that factors such as the duration of follow-up and the selection criteria in randomised trials may exclude participants at an increased risk of harm.

Adverse events are seldom specified as primary outcomes or are not pre-specified at all, so there is often a lack of clarity in the methods used to obtain adverse events data. The trials included in this review employed varied approaches to define, monitor, and report adverse events. Some trials coded composite adverse and serious adverse events in a systematic way using the Medical Dictionary for Regulatory Activities (MeDRA) [33,36]. In one study, composite and serious adverse events were provided, along with the criteria used for their definition [37], while another provided only data on specific adverse events, leaving uncertainty about the occurrence of other adverse events [42]. Some trials reported several categories of adverse events without providing clear definition of each [34,35], others reported only total adverse events [38–40], and one trial did not provide any data on adverse events [41].

Adverse events may not necessarily be linked to the intervention product, even when they are known to be associated with that category of drug. Some of the included trials specifically addressed causality between adverse events and study drugs by presenting data on both adverse events observed regardless of relation to the study drug, and adverse events possibly, probably, or definitely related to the study drug [34,35,40], while other trials addressed this issue but did not provide such data [37], or they did not address this issue at all [33,38,39,41,42].

Most of the included trials presented all observed adverse events, while bigger trials presented the most frequently occurring adverse events [33,35]. Finally, none of the included studies were designed to address safety as the primary outcome. Furthermore, we also decided to include reported symptoms related to drug administration when they were not specifically labelled as adverse events, in order not to potentially dismiss good-quality data due to a lack of correct phrasing. Finally, we performed meta-analyses of both composite and specific adverse events [47], though we are aware of the difficulties in interpreting composite adverse outcomes that are potentially constructed from several diverse events, since an important signal of rare serious adverse events could be masked by common trial adverse events [26].

Our study is based solely on published data due to practical constraints, acknowledging that information taken from published reports may be incomplete or lack specificity [48]. This limitation arises from challenges in accessing unpublished information, introducing the possibility of discordant findings. Of note, our analysis did not reveal signs of publication bias, suggesting that the observed reduction in thromboembolic events for the fibrinogen arm was not disproportionately influenced by selectively reported positive outcomes. While the exclusion of non-published data is a limitation, the absence of publication bias adds credibility to our findings, emphasizing the need for future research to address this limitation and enhance the overall robustness of conclusions in this field. To the best of our knowledge, a search of trial registries yielded no ongoing studies assessing safety

outcomes in non-obstetric and non-trauma adult patients who underwent cardiovascular, abdominal, or orthopaedic surgery and received fibrinogen replacement during the perioperative phase.

Our data showed that the prophylactic use of fibrinogen resulted in a reduction in ICU stay duration compared to the therapeutic use of fibrinogen. However, it is crucial to approach these findings with caution, due to the inherent complexities in assessing ICU stay as a safety indicator. Patient characteristics such as age and pre-existing health conditions, variations in clinical practices, and the nature of surgical procedures can all influence the interpretation of these results. Additionally, the impact of evolving medical practices over time underscores the need for a nuanced understanding when evaluating the safety implications of fibrinogen use in perioperative care.

Readers of this review should therefore be mindful of these limitations when critically appraising our data. It is important to remember that association does not imply causality, particularly considering the multitude of factors that contribute to adverse events and outcomes.

This study has numerous strengths. Our review strictly followed a predefined methodology from the Cochrane Handbook for Systematic Reviews of Interventions and was conducted using a previously registered protocol. Additionally, we employed an extensive search strategy using leading bibliographic databases for biomedical research. Unlike other reviews on the topic, we focused exclusively on a specific group of patients, thereby reducing potential bias and the heterogeneity that can result from varied population characteristics.

We acknowledge certain limitations to our study. Some of the included studies had small sample sizes, which may limit the ability to detect rare adverse events. We found a significant number of studies that report on safety outcomes after FC administration, but considerable heterogeneity remains. This includes variances in the countries where the studies took place, data being mainly drawn from cardiac surgery studies, different administration methods adhering to various fibrinogen level thresholds or ROTEM FIBTEM care test targets, and the diverse dosages of FC in all the trials. Such variances could potentially influence the broad applicability and interpretation of our results. Furthermore, the relevance of findings from older studies included in our research could be questionable, due to evolving standards of care and blood management protocols.

In this study, the identified trials comprised patients predominantly undergoing cardiac surgery, but also abdominal and orthopaedic surgeries. We recognise that the inclusion of different types of surgery increases heterogeneity (different clinical contexts, triggers for fibrinogen supplementation, doses of fibrinogen concentrate, and target values), and, due to the limited number of available studies, the applicability of these study findings to other surgical interventions is uncertain. Nevertheless, at a certain point, all surgical patients developed hypofibrinogenaemia, necessitating fibrinogen replacement to control coagulopathy, which may allow for the generalisation of these findings. Thus, the perioperative period can be viewed as a major group of patients, while trauma and postpartum haemorrhage have pathophysiological mechanisms that may imply different triggers and target values for fibrinogen supplementation and/or other haemostatic agents.

We tried to overcome the heterogeneity of the included studies by conducting a sensitivity analysis focusing exclusively on cardiovascular surgery studies, given their predominance within the included trials. However, despite the specificity of the patient population undergoing cardiovascular surgery, fibrinogen administration did not yield significant differences in the risk of total thromboembolic events, overall mortality, length of hospital stays, or any other safety outcomes compared to the control groups. These findings suggest that fibrinogen's effects may vary substantially across different surgical populations and underscore the need for continued research to elucidate its precise role in clinical practice. Furthermore, we conducted subgroup analyses in order to assess the impact of different comparators and different regimens. However, as the number of included studies is low, the results of these analyses should be interpreted with caution, as the analyses will lack statistical power.

No published systematic reviews with similar results to our current study have been identified. Fominskiy et al. [16] noted a decrease in overall mortality in the FC group, with only four trials contributing to the analysis [34,35,37,40], but detected no difference in the incidence of thrombotic events. A recent review by Ka Ting and associates [15] did not assess mortality and found no significant variations in thromboembolic events using data from only three trials.

In evaluating the safety of any intervention, the assessment of mortality should be considered a mandatory aspect. Despite the possible controversy surrounding mortality as a primary outcome, it is still a crucial safety measure, as it encapsulates both the ultimate harms and benefits of a treatment. Even if participants in the trials included may represent patients with generally low mortality rates, the consideration of this outcome holds significant clinical importance, especially when extrapolating the data to scenarios involving severe, acute bleeding—a common situation in everyday medical practice. Nevertheless, we acknowledge the need for caution when interpreting mortality results from clinical trials. Additionally, not all studies included mortality as a primary endpoint.

Importantly, it is essential to recognise the difficulty in associating a death event solely with an intervention, without considering various contributing factors such as clinical history, surgery risk, age, and complications during and after the operative period. Unfortunately, many studies fail to report whether such events were likely or definitively related to the outcome.

Indeed, the absence of consensus among various published reviews can be attributed to differences in methodologies, clinical settings, age ranges, operational timings, and the reported results [14–16,19–21]. Additionally, the consideration of two new trials involving 780 patients, which were not included in prior reviews, highlights the ever-changing nature of evidence in this field [33,36].

Although this review features recent studies with larger sample sizes and refined designs, there is still a pressing need for additional trials with well-defined groups of patients. It is crucial to focus on minimising bias and amplifying statistical potency to highlight differences in clinically significant patient safety measures. Extensive multicentre trials during the perioperative period are required to validate the effects of FC on these parameters.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/jcm13123482/s1, Table S1: Preferred Reporting Items for Systematic Reviews and Meta-Analyses checklist; Table S2: Retrieval search strategy; Table S3: Summary of effect measures for fibrinogen concentrate administration outcomes in cardiovascular surgery studies; Figure S1: Forest plots for stroke or TIA according to comparator group. TIA, transient ischaemic attack; Figure S2: Forest plots for myocardial infarction according to comparator group; Figure S3: Forest plots for PE or DVT according to comparator group. PE, pulmonary embolism; DVT, deep venous thrombosis; Figure S4: Forest plots for stroke or TIA according to indication setting. TIA, transient ischaemic attack; Figure S5: Forest plots for myocardial infarction according to indication setting; Figure S6: Forest plots for PE or DVT according to indication setting. PE, pulmonary embolism; DVT, deep venous thrombosis; Figure S7: Forest plots for mortality according to indication setting; Figure S8: Forest plots for hospitalisation time according to comparator group; Figure S9: Forest plots for ICU time according to comparator group; Figure S10: Forest plots for serious adverse events according to comparator group; Figure S11: Forest plots for adverse events according to indication setting; Figure S12: Forest plots for serious adverse events according to indication setting.

Author Contributions: M.G.: Conceptualisation, Investigation, Methodology, Data curation, Validation, Writing—review and editing. M.Â.-D.: Investigation, Methodology, Data curation Validation, Writing—Original Draft Preparation. G.S.D.: Methodology, Data curation, Validation, Formal analysis, Resources, Writing—review and editing. S.S.D.: Methodology, Validation, Formal analysis, Resources. S.S.S.: Investigation, Methodology, Data curation, Resources. J.L.: Conceptualisation, Methodology, Validation, Resources, Writing—review and editing, Supervision, Project administration. All authors were involved in the critical revision of the manuscript. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: No ethics committee approval was required because the study analysed only published data.

Acknowledgments: The authors thank CSL Behring for the article processing fee (APC) for the manuscript.

Conflicts of Interest: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

References

1. Kauvar, D.S.; Lefering, R.; Wade, C.E. Impact of Hemorrhage on Trauma Outcome: An Overview of Epidemiology, Clinical Presentations, and Therapeutic Considerations. *J. Trauma Inj. Infect. Crit. Care* **2006**, *60*, S3–S11. [CrossRef] [PubMed]
2. Hiippala, S.T.; Myllyla, G.J.; Vahtera, E.M. Hemostatic factors and replacement of major blood loss with plasma-poor red cell concentrates. *Anesth. Analg.* **1995**, *81*, 360–365. [PubMed]
3. Charbit, B.; Mandelbrot, L.; Samain, E.; Baron, G.; Haddaoui, B.; Keita, H.; Sibony, O.; Hurtaud-Roux, M.F.; Huisse, M.G.; Denninger, M.H.; et al. The decrease of fibrinogen is an early predictor of the severity of postpartum hemorrhage. *J. Thromb. Haemost.* **2007**, *5*, 266–273. [CrossRef] [PubMed]
4. Stinger, H.K.; Spinella, P.C.; Perkins, J.G.; Grathwohl, K.W.; Salinas, J.; Martini, W.Z.; Hess, J.R.; Dubick, M.A.; Simon, C.D.; Beekley, A.C.; et al. The Ratio of Fibrinogen to Red Cells Transfused Affects Survival in Casualties Receiving Massive Transfusions at an Army Combat Support Hospital. *J. Trauma Inj. Infect. Crit. Care* **2008**, *64*, S79–S85; discussion S85. [CrossRef] [PubMed]
5. Rossaint, R.; Afshari, A.; Bouillon, B.; Cerny, V.; Cimpoesu, D.; Curry, N.; Duranteau, J.; Filipescu, D.; Grottke, O.; Grønlykke, L.; et al. The European guideline on management of major bleeding and coagulopathy following trauma: Sixth edition. *Crit. Care* **2023**, *27*, 80. [CrossRef] [PubMed]
6. Levy, J.H.; Welsby, I.; Goodnough, L.T. Fibrinogen as a therapeutic target for bleeding: A review of critical levels and replacement therapy. *Transfusion* **2014**, *54*, 1389–1405. [CrossRef] [PubMed]
7. Besser, M.W.; MacDonald, S.G. Acquired hypofibrinogenemia: Current perspectives. *J. Blood Med.* **2016**, *7*, 217–225. [CrossRef] [PubMed]
8. Levy, J.H.; Szlam, F.; Tanaka, K.A.; Sniecienski, R.M. Fibrinogen and hemostasis: A primary hemostatic target for the management of acquired bleeding. *Anesth. Analg.* **2012**, *114*, 261–274. [CrossRef] [PubMed]
9. Costa-Filho, R.; Hochleitner, G.; Wendt, M.; Teruya, A.; Spahn, D.R. Over 50 Years of Fibrinogen Concentrate. *Clin. Appl. Thromb./Hemost.* **2016**, *22*, 109–114. [CrossRef] [PubMed]
10. Kozek-Langenecker, S. Clinical efficacy of fresh frozen plasma compared with coagulation factor concentrates for treating coagulopathy in patients with massive bleeding. *Med. Intensiv.* **2016**, *40*, 371–373. [CrossRef]
11. Levy, J.H.; Grottke, O.; Fries, D.; Kozek-Langenecker, S. Therapeutic Plasma Transfusion in Bleeding Patients: A Systematic Review. *Obstet. Anesthesia Dig.* **2017**, *124*, 1268–1276. [CrossRef] [PubMed]
12. Carvalho, M.; Rodrigues, A.; Gomes, M.; Carrilho, A.; Nunes, A.R.; Orfão, R.; Alves, Â.; Aguiar, J.; Campos, M. Interventional Algorithms for the Control of Coagulopathic Bleeding in Surgical, Trauma, and Postpartum Settings: Recommendations From the Share Network Group. *Clin. Appl. Thromb.* **2016**, *22*, 121–137. [CrossRef] [PubMed]
13. Kietaibl, S.; Ahmed, A.; Afshari, A.; Albaladejo, P.; Aldecoa, C.; Barauskas, G.; De Robertis, E.; Faraoni, D.; Filipescu, D.C.; Fries, D.; et al. Management of severe peri-operative bleeding: Guidelines from the European Society of Anaesthesiology and Intensive Care: Second update 2022. *Eur. J. Anaesthesiol.* **2023**, *40*, 226–304. [PubMed]
14. Wikkelsø, A.; Lunde, J.; Johansen, M.; Stensballe, J.; Wetterslev, J.; Møller, A.M.; Afshari, A. Fibrinogen concentrate in bleeding patients. *Cochrane Database Syst. Rev.* **2013**, *2013*, CD008864. [CrossRef] [PubMed]
15. Ng, K.T.; Yap, J.L.L.; Kwok, P.E. The effect of fibrinogen concentrate on postoperative blood loss: A systematic review and meta-analysis of randomized controlled trials. *J. Clin. Anesth.* **2020**, *63*, 109782. [CrossRef] [PubMed]
16. Fominskiy, E.; Nepomniashchikh, V.A.; Lomivorotov, V.V.; Monaco, F.; Vitiello, C.; Zangrillo, A.; Landoni, G. Efficacy and Safety of Fibrinogen Concentrate in Surgical Patients: A Meta-Analysis of Randomized Controlled Trials. *J. Cardiothorac. Vasc. Anesthesia* **2016**, *30*, 1196–1204. [CrossRef] [PubMed]
17. Appel, I.M.; Grimminck, B.; Geerts, J.; Stigter, R.; Cnossen, M.H.; Beishuizen, A. Age dependency of coagulation parameters during childhood and puberty. *J. Thromb. Haemost.* **2012**, *10*, 2254–2263. [CrossRef] [PubMed]
18. Davenport, R. Pathogenesis of acute traumatic coagulopathy. *Transfusion* **2013**, *53* (Suppl. S1), 23S–27S. [CrossRef] [PubMed]
19. Warmuth, M.; Mad, P.; Wild, C. Systematic review of the efficacy and safety of fibrinogen concentrate substitution in adults. *Acta Anaesthesiol. Scand.* **2012**, *56*, 539–548. [CrossRef] [PubMed]
20. Itagaki, Y.; Hayakawa, M.; Takahashi, Y.; Hirano, S.; Yamakawa, K. Emergency administration of fibrinogen concentrate for haemorrhage: Systematic review and meta-analysis. *World J. Emerg. Surg.* **2023**, *18*, 27. [CrossRef] [PubMed]
21. Stabler, S.N.B.; Li, S.S.; Karpov, A.M.; Vu, E.N.M. Use of fibrinogen concentrate for trauma-related bleeding: A systematic-review and meta-analysis. *J. Trauma Acute Care Surg.* **2020**, *89*, 1212–1224. [CrossRef] [PubMed]
22. Page, M.J.; McKenzie, J.E.; Bossuyt, P.M.; Boutron, I.; Hoffmann, T.C.; Mulrow, C.D.; Shamseer, L.; Tetzlaff, J.M.; Akl, E.A.; Brennan, S.E.; et al. The PRISMA 2020 statement: An updated guideline for reporting systematic reviews. *Int. J. Surg.* **2021**, *88*, 105906. [CrossRef] [PubMed]

23. Page, M.J.; Moher, D.; Bossuyt, P.M.; Boutron, I.; Hoffmann, T.C.; Mulrow, C.D.; Shamseer, L.; Tetzlaff, J.M.; Akl, E.A.; Brennan, S.E.; et al. PRISMA 2020 explanation and elaboration: Updated guidance and exemplars for reporting systematic reviews. *BMJ* **2021**, *372*, n160. [CrossRef] [PubMed]
24. Ouzzani, M.; Hammady, H.; Fedorowicz, Z.; Elmagarmid, A. Rayyan—A web and mobile app for systematic reviews. *Syst. Rev.* **2016**, *5*, 210. [CrossRef] [PubMed]
25. Microsoft Corporation. Microsoft Excel. 2018. Available online: https://office.microsoft.com/excel (accessed on 1 February 2024).
26. Peryer, G.; Golder, S.; Junqueira, D.; Vohra, S.; Loke, Y.K. Chapter 19: Adverse effects. In *Cochrane Handbook for Systematic Reviews of Interventions Version 6.4*; Updated August, 2023; Higgins, J.P.T., Thomas, J., Chandler, J., Cumpston, M., Li, T., Page, M.J., Welch, V.A., Eds.; Cochrane: London, UK, 2023. Available online: https://www.training.cochrane.org/handbook/current/chapter-09 (accessed on 1 February 2024).
27. Higgins, J.P.T.; Savović, J.; Page, M.J.; Elbers, R.G.; Sterne, J.A.C. Chapter 8: Assessing risk of bias in a randomized trial. In *Cochrane Handbook for Systematic Reviews of Interventions Version 6.3*; Updated February, 2022; Higgins, J.P.T., Thomas, J., Chandler, J., Cumpston, M., Li, T., Page, M.J., Welch, V.A., Eds.; Cochrane: London, UK, 2022. Available online: https://www.training.cochrane.org/handbook/current/chapter-08 (accessed on 1 February 2024).
28. Higgins, J.P.; Altman, D.G.; Gøtzsche, P.C.; Jüni, P.; Moher, D.; Oxman, A.D.; Savović, J.; Schulz, K.F.; Weeks, L.; Sterne, J.A. The Cochrane Collaboration's tool for assessing risk of bias in randomised trials. *BMJ* **2011**, *343*, d5928. [CrossRef] [PubMed]
29. Viechtbauer, W. Conducting Meta-Analyses in R with the metafor Package. *J. Stat. Softw.* **2010**, *36*, 1–48. [CrossRef]
30. Higgins, J.P.T.; Li, T.; Deeks, J.J. Chapter 6: Choosing effect measures and computing estimates of effect. In *Cochrane Handbook for Systematic Reviews of Interventions Version 6.4*; Updated August, 2023; Higgins, J.P.T., Thomas, J., Chandler, J., Cumpston, M., Li, T., Page, M.J., Welch, V.A., Eds.; Cochrane: London, UK, 2023. Available online: https://www.training.cochrane.org/handbook/current/chapter-06 (accessed on 1 February 2024).
31. Higgins, J.P.T.; Thompson, S.G.; Deeks, J.J.; Altman, D.G. Measuring inconsistency in meta-analyses. *BMJ* **2003**, *327*, 557–560. [CrossRef] [PubMed]
32. Page, M.J.; Higgins, J.P.T.; Sterne, J.A.C. Chapter 13: Assessing risk of bias due to missing results in a synthesis. In *Cochrane Handbook for Systematic Reviews of Interventions Version 6.4*; Updated August, 2023; Higgins, J.P.T., Thomas, J., Chandler, J., Cumpston, M., Li, T., Page, M.J., Welch, V.A., Eds.; Cochrane: London, UK, 2023. Available online: https://www.training.cochrane.org/handbook/current/chapter-13 (accessed on 1 February 2024).
33. Callum, J.; Farkouh, M.E.; Scales, D.C.; Heddle, N.M.; Crowther, M.; Rao, V.; Hucke, H.P.; Carroll, J.; Grewal, D.; Brar, S.; et al. Effect of Fibrinogen Concentrate vs Cryoprecipitate on Blood Component Transfusion After Cardiac Surgery: The FIBRES Randomized Clinical Trial. *JAMA—J. Am. Med. Assoc.* **2019**, *322*, 1966–1976. [CrossRef] [PubMed]
34. Rahe-Meyer, N.; Solomon, C.; Hanke, A.; Schmidt, D.S.; Knoerzer, D.; Hochleitner, G.; Sørensen, B.; Hagl, C.; Pichlmaier, M. Effects of fibrinogen concentrate as first-line therapy during major aortic replacement surgery: A randomized, placebo-controlled trial. *Anesthesiology* **2013**, *118*, 40–50. [CrossRef] [PubMed]
35. Rahe-Meyer, N.; Levy, J.H.; Mazer, C.D.; Schramko, A.; Klein, A.A.; Brat, R.; Okita, Y.; Ueda, Y.; Schmidt, D.S.; Ranganath, R.; et al. Randomized evaluation of fibrinogen vs placebo in complex cardiovascular surgery (REPLACE): A double-blind phase III study of haemostatic therapy. *Br. J. Anaesth.* **2016**, *117*, 41–51. [CrossRef] [PubMed]
36. Roy, A.; Stanford, S.; Nunn, S.; Alves, S.; Sargant, N.; Rangarajan, S.; Smith, E.A.; Bell, J.; Dayal, S.; Cecil, T.; et al. Efficacy of fibrinogen concentrate in major abdominal surgery—A prospective, randomized, controlled study in cytoreductive surgery for pseudomyxoma peritonei. *J. Thromb. Haemost.* **2020**, *18*, 352–363. [CrossRef] [PubMed]
37. Sabate, A.; Gutierrez, R.; Beltran, J.; Mellado, P.; Blasi, A.; Acosta, F.; Costa, M.; Reyes, R.; Torres, F. Impact of Preemptive Fibrinogen Concentrate on Transfusion Requirements in Liver Transplantation: A Multicenter, Randomized, Double-Blind, Placebo-Controlled Trial. *Am. J. Transplant.* **2016**, *16*, 2421–2429. [CrossRef] [PubMed]
38. Najafi, A.; Moharari, R.S.; Orandi, A.A.; Etezadi, F.; Sanatkar, M.; Khajavi, M.R.; Ahmadi, A.; Pourfakhr, P.; Imani, F.; Mojtahedzadeh, M.; et al. Prophylactic administration of fibrinogen concentrate in perioperative period of total hip arthroplasty: A randomized clinical trial study. *Acta Medica Iran.* **2014**, *52*, 804–810.
39. Karlsson, M.; Ternström, L.; Hyllner, M.; Baghaei, F.; Flinck, A.; Skrtic, S.; Jeppsson, A. Prophylactic fibrinogen infusion reduces bleeding after coronary artery bypass surgery. A prospective randomised pilot study. *Thromb. Haemost.* **2009**, *102*, 1137–1144. [CrossRef] [PubMed]
40. Ranucci, M.; Baryshnikova, E.; Crapelli, G.B.; Rahe-Meyer, N.; Menicanti, L.; Frigiola, A.; Surgical Clinical Outcome REsearch (SCORE) Group. Randomized, double-blinded, placebo-controlled trial of fibrinogen concentrate supplementation after complex cardiac surgery. *J. Am. Heart Assoc.* **2015**, *4*, e002066. [CrossRef] [PubMed]
41. Tanaka, K.A.; Egan, K.; Szlam, F.; Ogawa, S.; Roback, J.D.; Sreeram, G.; Guyton, R.A.; Chen, E.P. Transfusion and hematologic variables after fibrinogen or platelet transfusion in valve replacement surgery: Preliminary data of purified lyophilized human fibrinogen concentrate versus conventional transfusion. *Transfusion* **2014**, *54*, 109–118. [CrossRef] [PubMed]
42. Bilecen, S.; de Groot, J.A.; Kalkman, C.J.; Spanjersberg, A.J.; Bruinsma, G.J.B.B.; Moons, K.G.; Nierich, A.P. Effect of Fibrinogen Concentrate on Intraoperative Blood Loss Among Patients with Intraoperative Bleeding during High-Risk Cardiac Surgery: A Randomized Clinical Trial. *JAMA—J. Am. Med. Assoc.* **2017**, *317*, 738–747. [CrossRef] [PubMed]

43. Ferko, N.; Corral, M.; Hollmann, S.; Broder, M.S.; Chang, E. Health and economic outcomes associated with uncontrolled surgical bleeding: A retrospective analysis of the Premier Perspectives Database. *Clin. Outcomes Res.* **2015**, *7*, 409–421. [CrossRef] [PubMed]
44. Guzzetta, N.A.; Allen, N.N.; Wilson, E.C.; Foster, G.S.; Ehrlich, A.C.; Miller, B.E. Excessive Postoperative Bleeding and Outcomes in Neonates Undergoing Cardiopulmonary Bypass. *Obstet. Anesthesia Dig.* **2015**, *120*, 405–410. [CrossRef] [PubMed]
45. Ponschab, M.; Landoni, G.; Biondi-Zoccai, G.; Bignami, E.; Frati, E.; Nicolotti, D.; Monaco, F.; Pappalardo, F.; Zangrillo, A. Recombinant Activated Factor VII Increases Stroke in Cardiac Surgery: A Meta-analysis. *J. Cardiothorac. Vasc. Anesth.* **2011**, *25*, 804–810. [CrossRef] [PubMed]
46. Fergusson, D.A.; Hébert, P.C.; Mazer, C.D.; Fremes, S.; MacAdams, C.; Murkin, J.M.; Teoh, K.; Duke, P.C.; Arellano, R.; Blajchman, M.A.; et al. A Comparison of Aprotinin and Lysine Analogues in High-Risk Cardiac Surgery. *N. Engl. J. Med.* **2008**, *358*, 2319–2331. [CrossRef] [PubMed]
47. Choi, S.W.; Lam, D.M.H. Trials and tribulations of a meta-analyst. *Anaesthesia* **2016**, *71*, 228–231. [CrossRef] [PubMed]
48. Golder, S.; Loke, Y.K.; Wright, K.; Norman, G. Reporting of Adverse Events in Published and Unpublished Studies of Health Care Interventions: A Systematic Review. *PLoS Med.* **2016**, *13*, e1002127. [CrossRef] [PubMed]

CHAPTER IV

Safety of Fibrinogen Concentrate for Correcting Perioperative Bleeding-Associated Hypofibrinogenemia in Adults: A Single-Center Experience

Journal of
Clinical Medicine

MDPI

Article

Safety of Fibrinogen Concentrate for Correcting Perioperative Bleeding-Associated Hypofibrinogenemia in Adults: A Single-Center Experience

Manuela Gomes [1], Miguel Ângelo-Dias [2,3] and Jorge Lima [2,3,4,*]

[1] Transfusion Medicine Department, Hemovida, Hospital da Luz Lisboa, Luz Saúde, 1500-650 Lisboa, Portugal; amdrgv@gmail.com
[2] NOVA Medical School, Faculdade de Ciências Médicas, NMS, FCM, Universidade Nova de Lisboa, 1169-056 Lisboa, Portugal; miguel.dias@nms.unl.pt
[3] CHRC, NOVA Medical School, Faculdade de Ciências Médicas, NMS, FCM, Universidade Nova de Lisboa, 1169-056 Lisboa, Portugal
[4] Department of Obstetrics and Gynecology, Hospital da Luz Lisboa, Luz Saúde, Avenida Lusíada 100, 1500-650 Lisboa, Portugal
* Correspondence: jorgeramoslima@sapo.pt

Abstract: Background: Surgery often leads to bleeding associated with hypofibrinogenemia. Supplementation with fibrinogen concentrate appears to be effective and safe, although findings from studies are inconsistent. The primary aim of this study was to assess the safety of fibrinogen concentrate during the perioperative period. **Methods:** This single-centre, prospective, observational study included adult patients undergoing scheduled or emergency surgery related to bleeding coagulopathy and the administration of fibrinogen concentrate. Patients were followed until their discharge from the institution. Comprehensive data were collected, including age, sex, type of surgery, associated comorbidities, anticoagulant and/or anti-aggregating therapy, and the number of blood transfusions. Laboratory data on plasma fibrinogen concentration, haemoglobin, and platelet count before and after surgery were also collected. The primary outcomes were the mortality rate at discharge and any reported thrombotic or thromboembolic events, including deep vein thrombosis, pulmonary embolism, and myocardial infarction. **Results:** The study included 91 adult patients who had undergone surgery, with 29 surgeries (32%) conducted in an emergency setting. The mean age was 59.2 years, and 53.8% were male. Major bleeding occurred in 29 cases, mainly in older males and those on anticoagulant therapy. The pre-operative fibrinogen level averaged 161 mg/dL, and the average dosage of fibrinogen concentrate administered was 2.7 g. Eight patients died (8.8%), mostly due to septic or cardiogenic shock, with deaths being more frequent in emergency settings. Thromboembolic events occurred in eight patients, none of whom died. No additional adverse events directly related to the administration of fibrinogen concentrate were reported. **Conclusions:** Our findings suggest a favourable safety profile for fibrinogen concentrate in surgical patients, as evidenced by a low incidence of deaths and thromboembolic events, which were primarily attributed to other factors. Future research should strive to increase statistical robustness to further illuminate clinically significant patient safety measures.

Keywords: fibrinogen concentrate; perioperative care; hypofibrinogenemia; thrombosis; haemostasis

check for updates

Citation: Gomes, M.; Ângelo-Dias, M.; Lima, J. Safety of Fibrinogen Concentrate for Correcting Perioperative Bleeding-Associated Hypofibrinogenemia in Adults: A Single-Center Experience. *J. Clin. Med.* **2024**, *13*, 6018. https://doi.org/10.3390/jcm13196018

Academic Editors: X. Long Zheng and Emmanuel Favaloro

Received: 14 August 2024
Revised: 2 October 2024
Accepted: 7 October 2024
Published: 9 October 2024

1. Introduction

Surgery often results in severe bleeding, necessitating precise and effective haemostatic management. Central to this process is fibrinogen, the main plasma protein coagulation factor acknowledged as an early participant in events leading to clot formation. Primarily recognized for its role as the fibrin precursor, it becomes cleaved by thrombin, and fibrinogen-derived fibrin monomers then polymerize to form a net that entraps cells,

including platelets, establishing the clot's foundation. Additionally, fibrinogen significantly contributes to platelet aggregation by binding to GP IIb/IIIa receptors on activated platelets. Furthermore, as an acute-phase reactant, fibrinogen modulates inflammatory cellular responses, underscoring its crucial role in maintaining primary and secondary haemostasis [1,2].

Synthesized in hepatocytes, fibrinogen's plasma concentration typically ranges from 1.5 to 4.5 g/L. However, in circumstances such as pregnancy, these levels can become even higher [1,3]. Various factors influence normal fibrinogen levels, including physiological (age, sex), pathological (hepatic disease), and lifestyle factors (smoking) [1]. Conversely, bleeding episodes can result in fibrinogen loss, dilution, and consumption, compounded further by factors such as hypothermia and acidosis, which can impair its function [3,4]. Therefore, patients experiencing significant bleeding during surgery are at an increased risk of developing hypofibrinogenemia, where fibrinogen levels drop to critically low levels.

The best approach to fibrinogen supplementation remains under debate, with three main methods: plasma, cryoprecipitate, and fibrinogen concentrate.

Plasma is not the optimal source for fibrinogen replacement. There are various types of plasma, namely solvent detergent plasma and fresh frozen plasma (FFP), with fibrinogen levels typically between 2 and 2.9 g/L. Some FFP units may have levels as low as 1 g/L. As a result, substantial volumes are necessary to achieve a substantial correction in fibrinogen levels. Additionally, plasma needs to be ABO-compatible and thawed, necessitating preparation time and equipment [5,6]. Cryoprecipitate, while containing higher fibrinogen levels than FFP, has an uncertain fibrinogen concentration and also contains other coagulation factors. It is usually administered as a pooled product originating from 6 to 10 units of blood, increasing the recipient's exposure to multiple donors. The primary concern with cryoprecipitate is its safety profile since it does not undergo antiviral processing and is not available for use in several European countries due to safety concerns [4].

Fibrinogen concentrate is a rigorously purified, pathogen-inactivated, and lyophilized product derived from plasma. This results in a sterile, stable product that provides a pre-determined and standardized amount of fibrinogen replacement per vial. Given these qualities, fibrinogen concentrate has become a go-to intervention for correcting hypofibrinogenemia-associated coagulopathy during the perioperative period. It allows for accurate and quick restoration of fibrinogen levels, thus promoting successful clot formation and platelet function. Pharmacovigilance data accumulated over the decades suggest that fibrinogen concentrate supplementation, in cases of congenital and acquired fibrinogen deficiency, does not lead to adverse events [7,8]. However, the trigger and target levels for fibrinogen supplementation vary and appear to depend on multiple factors. That said, several guidelines propose that fibrinogen levels between 1.5 and 2.0 g/L should be adjusted via supplementation [9,10].

Although numerous studies have reviewed the use of fibrinogen concentrate in correcting hypofibrinogenemia and improving haemostasis [7,11–14], further investigation is needed to thoroughly assess its efficiency and safety profile. A significant concern is that fibrinogen concentrate, a procoagulant, is administered to patients already posing risk factors for thrombotic or thromboembolic events. While most clinical trials suggest fibrinogen concentrate's safety and effectiveness, inconsistencies in study designs, small sample sizes, varying institutional protocols, pre-existing conditions and medicines in patients, imprecise triggers and targets for fibrinogen supplementation, and a variation in dosages, often unrepeated in complicated and major surgeries, contribute to inconsistent findings. Moreover, the clinical and lab-based decisions on when and how to supplement fibrinogen vary widely. The conflicting data in the existing literature emphasize the need for a rigorous evaluation of the efficiency and safety of administering fibrinogen concentrate in the perioperative setting [13–23].

The primary objective of this study was to assess the safety of fibrinogen concentrate when administered within therapeutic ranges during the perioperative period to correct bleeding-associated hypofibrinogenemia in adults. Conducting a real-world analysis, this

research aimed to provide valuable insights into the practical use and safety profile of fibrinogen concentrate, addressing a vital aspect of managing haemostasis in surgical and bleeding scenarios.

2. Materials and Methods

2.1. Study Design

This was a prospective observational study conducted at Hospital da Luz Lisboa, Portugal, between July 2022 and December 2023. The study included all adult patients (≥18 years of age) who underwent scheduled or emergency surgery and required the administration of fibrinogen concentrate for correcting perioperative bleeding-associated hypofibrinogenemia. These patients were followed until their discharge from the institution. Fibrinogen concentrate doses (Haemocomplettan CSL Behring, Marburg, Germany) within the range of 25–50 mg/kg/dose were administered to all the patients in this study.

Patients with hereditary bleeding disorders that lead to fibrinogen deficiency or abnormal function were excluded. Patients exhibiting bleeding conditions accompanied by acquired hypofibrinogenemia associated with postpartum trauma, or any other medical condition, such as gastrointestinal bleeding, were also excluded.

2.2. Bleeding Protocol

The approach to managing bleeding patients adhered to our institution's protocol, grounded on the guidelines for major haemorrhages issued by the Portuguese Directorate of General Health [24]. This protocol underlines the importance of multidisciplinary communication and the critical initial steps in patient care. The essential initial steps comprise the following: the swift identification of surgical causes of bleeding and the implementation of necessary control measures; the reappraisal of the patient's therapeutic and medical history, particularly in emergency cases; the determination of haemoglobin and platelet levels; the assessment of coagulation status via standard coagulation screens or through the use of point-of-care test results (ROTEM® sigma—TEM Innovations, Munich, Germany); and the stabilization of the patient's basic physiological conditions, including addressing hypothermia, acidosis, hypocalcaemia, and anaemia.

This protocol outlines a sequential approach for addressing specific aspects of coagulopathy. Initially, if a clinical or laboratory suspicion of hyperfibrinolysis arises, correction with tranexamic acid is initiated at a dose of 20–25 mg/kg, followed by the administration of fibrinogen if a deficiency is observed, using fibrinogen concentrate (Haemocomplettan CSL Behring, 25–50 mg/kg). Thrombocytopenia is corrected with platelet concentrate, while a deficit of thrombin or other coagulation factors is addressed with prothrombin complex concentrate (PCC) (Octaplex Octapharma, 20–30 UI/kg) or FFP at a dose of 12–15 mL/kg. Treatments with haemostatic agents can be repeated as needed to control coagulopathy, always guided by standard coagulation tests and/or point-of-care test results and clinical evaluation.

Major bleeding was defined according to the volume of blood loss (one total blood volume (TBV)/24 h, or >50% of the TBV/3 h), ongoing bleeding rhythm (150 mL/min), and the number of transfused blood units.

The cut-off values considered for each transfused agent were as follows: RBC if <7 g/dL or 8 if heart disease; platelets if $<50 \times 10^9$/L and persistent active bleeding; FC if <200 mg/dL or blood loss > 1–1.5 L and ongoing bleeding, or according to ROTEM results; and PCC if international normalized ratio (INR) or activated partial thromboplastin time (aPTT) > 1.5 and acute active bleeding, or volume blood loss 150–200%, or 100% and liver disease. Repeated doses were applied depending on bleeding severity and the results of coagulation assessments after each treatment cycle.

Patients with anticoagulant or antiplatelet therapy were managed depending on the type of antithrombotic therapy, the time until surgery, and the risk of thromboembolic events. Namely, this occurred when surgery could not be delayed, and according to laboratory results, the procedure was as follows: for vitamin K antagonists, vitamin K and/or

PCC (15–30 IU/kg) were administered. For rapid reversal of direct oral anticoagulants, PCC was used at 25–50 IU/kg. In this group of patients, we had no need to use idarucizumab for dabigatran reversal. For antiplatelet therapy, a pool platelet concentrate was considered according to the clinical situation. Clinical indications for both therapies were formal indications, such as atrial fibrillation and coronary disease, among others.

The protocol was approved by the Institutional Ethics Committee, and informed consent was obtained from each study participant.

2.3. Data Collection

Data collection and patient follow-up began at the anaesthesiology appointment and concluded upon discharge from the hospital. Comprehensive data were gathered from each patient, including age, sex, type of surgery, associated comorbidities, anticoagulant and/or antiplatelet therapy, the number of red blood cell, and platelet concentrate units administered, as well as the total doses of fibrinogen concentrate, prothrombin complex concentrate, antithrombin concentrate, and FFP administered.

Additionally, when available, data regarding plasma fibrinogen concentration (determined by the Clauss method and/or results from viscoelastic testing), last haemoglobin, and platelet count before surgery, as well as the lowest haemoglobin and platelet count in the post-operative period were also collected.

The mortality rate at discharge, as well as all reported thrombotic or thromboembolic events, including deep vein thrombosis (DVT), pulmonary embolism (PE), myocardial infarction, and any other reported or clinically suspected thromboembolic event, were also recorded and documented. PE and DVT were monitored with ultrasound scan, D-Dimer Test, ECG chest X-ray, and clinical signs and symptoms. Most patients continued to be followed for a variable amount of time depending on personal residence and clinical outcome, but most of them were monitored for more than 3 months.

In addition, the presence of additional risk factors for thrombotic/thromboembolic events at the baseline, such as obesity, smoking status, family and past personal history of VTE, the use of oral contraceptives, hormone replacement therapy, and glucocorticoids, were also noted. Given that these factors might serve as confounding variables, these data facilitated further statistical stratification analysis.

2.4. Outcomes

The primary outcomes of interest in this study were overall mortality and the incidence of thromboembolic events at discharge. These events included myocardial infarction, DVT, PE, or stroke, all of which could potentially be associated with the administration of fibrinogen concentrate.

The secondary outcomes focused on additional adverse events reported, such as anaphylaxis, hypersensitivity, or allergic reactions, during the study period. They also focused on the total number of allogeneic blood components administered until discharge and the overall length of hospital stay.

2.5. Statistical Analysis

Categorical variables are presented as absolute frequencies and percentages; their associations were analysed using Fisher's exact test. Continuous data, if normally distributed, are expressed as the mean and standard deviation (SD); if not normally distributed, they are expressed as the median and interquartile range (IQR). The normality of these data was assessed by visually inspecting QQ plots of the residuals and by employing the D'Agostino–Pearson normality test when needed.

For data that are normally distributed, comparisons between two unpaired groups were conducted using the unpaired t-test or the unpaired t-test with Welch's correction, as appropriate. If not, the non-parametric Mann–Whitney test was employed. Correlations were determined using Spearman's correlation coefficient.

The test used in each case is specified in the respective figure and table legends. Any analysis with a *p*-value less than 0.05 was considered significant. Statistical analyses were conducted using GraphPad Prism v10.1.2 for Windows (GraphPad Software, Boston, MA, USA). Visualizations were created using Microsoft Power BI software v2.131.1203.0 for Windows (Microsoft Corporation, Redmond, WA, USA).

3. Results

3.1. Demographics, Comorbidities, and Concomitant Medication

A total of 91 adult patients were included in the study. Of these, 29 were urgent or emergency surgeries. Details of patient demographics and clinical characteristics are provided in Table 1.

Table 1. Demographic and clinical characteristics of all recruited patients.

	All Patients (n = 91)
Age, mean (SD), years	59.2 (18.2)
Males, n (%)	49 (53.8%)
Weight, mean (SD), kg	71.6 (15.3)
BMI, mean (SD), kg/m^2	25.1 (4.2)
Previous thromboembolic events, n (%)	11 (12.1%)
Family history of thromboembolic events, n (%)	3 (3.3%)
Smoking, n (%)	20 (22.0%)
Cancer, n (%)	15 (16.5%)
Autoimmune disease, n (%)	15 (16.5%)
Other comorbidities, n (%)	35 (61%)
Oral contraceptives, n (%)	9 (9.9%)
Hormone replacement therapy, n (%)	1 (1.1%)
Anticoagulant therapy, n (%)	16 (17.6%)
Anti-aggregating therapy, n (%)	20 (22.0%)
Glucocorticoid therapy, n (%)	3 (3.3%)
Hgb levels pre-op, mean (SD), g/dL	12.9 (2.1) [1]
Levels outside the normal range, n (%) [a]	48 (55.8%) [1]
Lowest Hgb levels post-op, mean (SD), g/dL	7.9 (1.6) [2]
Levels outside the normal range, n (%) [a]	89 (100%) [2]
Plt levels pre-op, median [min–max], $\times 10^9/L$	216 [69–833] [3]
Levels outside the normal range, n (%) [b]	18 (23.4%) [3]
Lowest Plt levels post-op, median [min–max], $\times 10^9/L$	139 [12–809] [4]
Levels outside the normal range, n (%) [b]	57 (71.3%) [4]
Plasma fibrinogen levels, mean (SD), mg/dL	161.0 (41.9) [5]
Levels outside the normal range, n (%) [c]	33 (91.7%) [5]
ROTEM MCF, mean (SD), mm	10.1 (2.8) [6]
Thromboembolic events, n (%) [d]	8 (8.8%)
Myocardial infarction	1
Deep vein thrombosis	2
Pulmonary embolism	2

Table 1. *Cont.*

	All Patients (n = 91)
Other	4
Mortality, n (%)	8 (8.8%)
No. of patients with platelet pool administered n (%)	26 (28.6%)
Units of Platelet pool administered, median [min–max]	0 [0–7]
No. of patients with prothrombin concentrate, n (%)	13 (14.3%)
Units of RBC administered, median [min–max]	2 [0–13]
No. of patients with FFP administered n (%)	36 (39.6%)
Units of FFP administered, median [min–max]	0 [0–8]
Doses of FC administered, mean (SD) [min–max], g	2.7 (1.4) [1–7]
Emergent surgery, n (%)	29 (31.9%)
Major bleeding, n (%)	29 (31.9%)
Type of surgery, n (%)	
Abdominal	33 (36.3%)
Cardiac/vascular	34 (37.4%)
Neurologic	10 (11.0%)
Orthopaedic	2 (2.2%)
Other	12 (13.1%)
Days of hospital stay, median [min–max] [7]	6 [1–80]

[a] Hgb normal range: 13.7–17.2 g/L; [b] Platelets normal range: 170–430 × 10^9/L; [c] Plasma fibrinogen normal range: 210–400 mg/dL; [d] Patients who experienced more than one event were counted only once in the total. [1] n = 86, [2] n = 89, [3] n = 77, [4] n = 80, [5] n = 36, [6] n = 24, [7] n = 83. SD, standard deviation; Hgb, haemoglobin; Plt, platelet; ROTEM, rotational thromboelastometry; MCF, maximum clot firmness; FFP, fresh frozen plasma; and FC, fibrinogen concentrate.

The mean age of the recruited patients was 59.2 ± 18.2 years, with an average weight of 71.6 ± 15.3 kg and a mean body mass index (BMI) of 25.1. Of these, forty-nine (53.8%) were males. Twenty patients were active smokers, and sixty-five had at least one related comorbidity. Before surgery, 36 patients were given medication that could potentially impair haemostasis, including anticoagulant and/or antiplatelet therapies.

Eleven patients (12.1%) had a history of previous thromboembolic events, and only three had a family history of such events.

3.2. Surgery and Bleeding

The types of surgery conducted in the study were predominantly cardiovascular (37%) or abdominal (36%) (Figure 1). Among the remaining surgeries, ten were spinal surgeries, two were orthopaedic procedures, and twelve, labelled as 'other', included thoracic and plastic surgeries.

Abdominal surgeries include procedures related to gynaecological, gastric, and prostatic issues, while cardiac surgeries primarily treat patients with valvular or coronary heart disease. The median length of hospital stay was 6 days, with a range of 1 to 80 days.

During the perioperative period, 29 major bleeding cases were documented, either during the intervention or within 48 h post-surgery (Figure 2). Of this cohort, 72% were male and older ($p = 0.014$), while 35% undertook anticoagulant therapy, received more units of RBC ($p < 0.001$), and fibrinogen concentrate ($p = 0.028$). Subsequently, they spent more days in the hospital ($p = 0.005$). Within these 29 patients, 11 patients received a platelet pool concentrate, 9 patients received PCC, 27 patients received RBC transfusions, and 14 patients received FFP. Only 5 patients were treated with all the aforementioned haemo-

static agents. Notably, nearly half of the patients subjected to emergent interventions experienced major bleeding ($p = 0.030$).

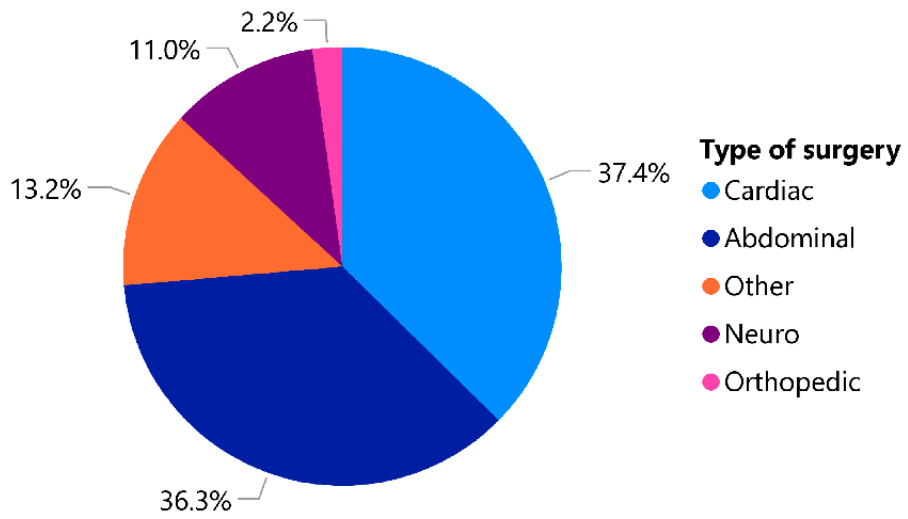

Figure 1. The proportions of patients by type of surgery.

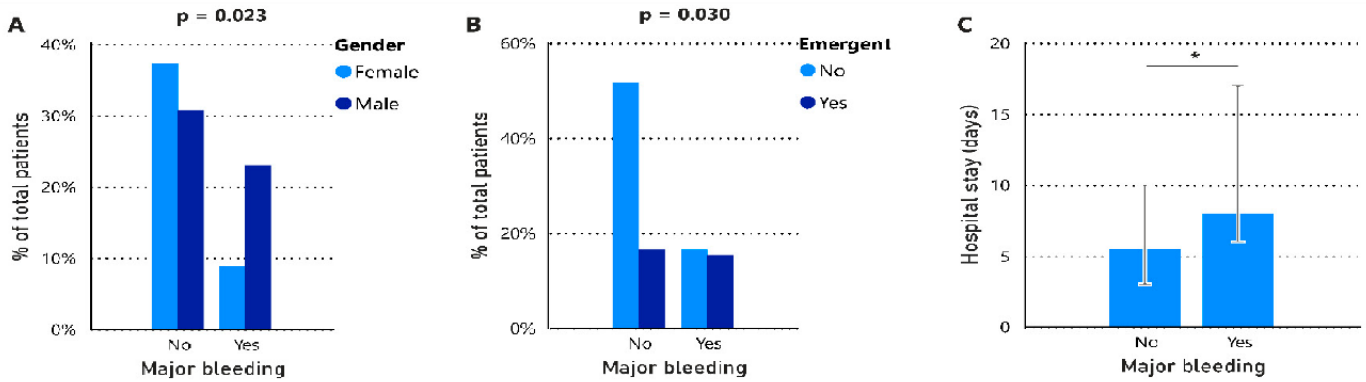

Figure 2. Distribution of patients by gender (**A**) and context of admission settings (**B**) according to the presence or absence of major bleeding. Bar plot showing with median with the IQR of the length of hospital stay for patients with and without major bleeding. Statistical analyses were performed using Fisher's exact test (**A**,**B**) or the Mann–Whitney test (**C**). IQR, interquartile range (25th–75th). * p-value < 0.05.

3.3. Perioperative Haemostasis and Blood Component Transfusions

The mean plasma fibrinogen levels, measured using the Clauss method, were 161 mg/dL at the decision point to administer fibrinogen concentrate, with five patients having levels below 100 mg/dL. Among patients who had point-of-care testing available at this time, seven had an FIBTEM MCF (Maximum Clot Firmness) below 9 mm, with the lowest value observed at 3 mm. The average dose of fibrinogen concentrate administered was 2.7 g, ranging from 1 to 7 g. All doses were calculated based on patient weight and fell within the recommended ranges, as previously described.

Before surgery, the mean haemoglobin level was measured at 12.9 ± 2.1 g/L, with the lowest post-operative level recorded at 7.9 ± 1.6 g/L. This represents an average decrease of 5 g/L. Similarly, the highest average pre-operative platelet level was 216 × 10^9/L, which fell to post-operative levels of 139 × 10^9/L, which is nearly a half. Following the hospital's bleeding management protocol, 26 patients received a platelet pool concentrate, 13 were given prothrombin concentrates, and 36 were administered FFP. These are detailed in Table 1.

Seventy-one patients received RBC transfusions, with a median of two units administered per patient. Among these, 20 patients received more than four units of RBC. Nineteen patients did not require any transfusions with blood components. These included three cardiovascular, five abdominal, five neurosurgical, one orthopaedic, and five thoracic surgeries, as bleeding was effectively managed using fibrinogen concentrate.

3.4. Primary Outcomes

3.4.1. Death

Eight patients from the study group succumbed following their surgeries, which translated to a mortality rate of 8.8%. The main causes of death were septic shock (n = 4), cardiogenic shock (n = 2), and hypovolemic shock (n = 2), all leading to multiple organ failure. The majority of deaths (seven of eight) occurred in patients who had undergone abdominal surgeries in contrast to only one resulting from cardiac surgery ($p = 0.027$). Of the patients who died, two received multiple transfusions, and none of the eight experienced any thromboembolic events. A significant association was found between the surgery setting and mortality (Figure 3), indicating a higher likelihood of deaths in emergent settings (20.7%) compared to non-emergent settings (3.2%). Furthermore, we noted that patients who passed away were older ($p = 0.037$), had lower fibrinogen levels at the time of the decision to administer fibrinogen concentrate ($p = 0.018$), and were recipients of multiple blood component transfusions. No significant variations were seen in the number of fibrinogen concentrate doses given between the two groups.

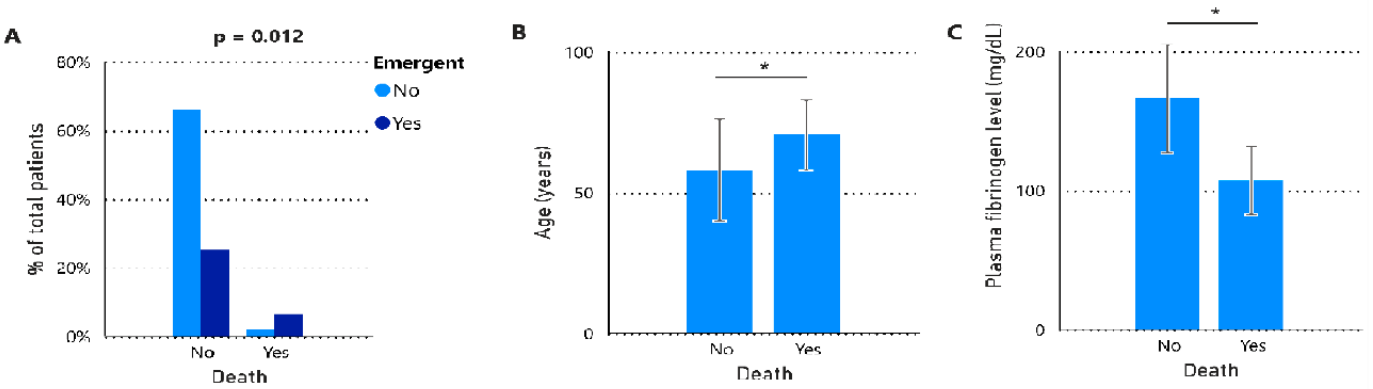

Figure 3. Distribution of patients by context of admission setting and death (**A**). Bar plots with mean ± SD of age (**B**) and plasma fibrinogen levels (**C**) according to the outcome of death. Statistical analyses were performed using Fisher's exact test (**A**), Unpaired *t*-test with Welch's correction (**B**), or unpaired *t*-test (**C**). SD, standard deviation. * *p*-value < 0.05.

3.4.2. Thromboembolic Events

Eight patients experienced at least one thromboembolic event post-surgery, with an instance of one patient enduring two such events (Figure 4). These occurrences included the following: one myocardial infarction, two deep vein thromboses, two pulmonary embolisms, and four strokes. Within this group, two patients also underwent multiple transfusions. Importantly, none of these patients died, and all were discharged from the hospital. Patients who experienced thromboembolic events received a greater quantity of RBC units ($p = 0.043$) and had lengthier median hospital stays before discharge ($p = 0.046$). In our research sample, half of the patients (n = 4) who manifested thromboembolic events had prior incidences of such events, in contrast to only 8.4% of the patients unaffected by such events (n = 7; $p = 0.007$). Only two patients experienced thromboembolic events and major bleeding. We observed no significant disparities in the fibrinogen concentrate doses administered across both groups.

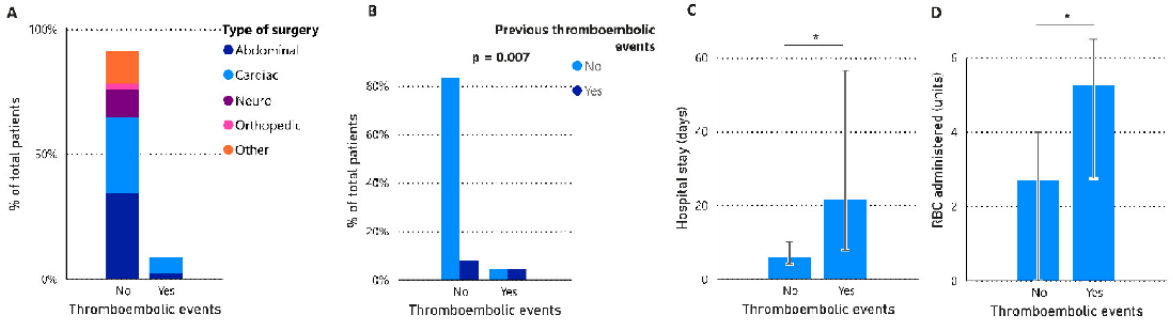

Figure 4. Distribution of patients by type of surgery (**A**) and history of previous thromboembolic events (**B**) according to the presence or absence of thromboembolic events during follow-up. Bar plots with median + IQR of the length of hospital stay (**C**) and units of RBC administered (**D**) in patients with and without thromboembolic events. Statistical analyses were performed using Fisher's exact test (**B**) or the Mann–Whitney test (**C**,**D**). IQR, interquartile range (25th–75th). * p-value < 0.05.

3.5. Secondary Outcomes

In this group of 91 patients, no adverse events directly associated with the administration of fibrinogen concentrate, such as anaphylaxis, hypersensitivity, or allergic reactions, were reported.

Older patients (over 65 years) typically necessitated longer hospital stays, with a median duration of 10 days compared to 5 days required for younger patients ($p < 0.001$). Moreover, age is inversely correlated with pre-surgery platelet levels (r = −0.253; $p = 0.026$). Conversely, age is positively correlated with the amount of RBC units administered (r = 0.281; $p = 0.007$) and the length of hospital stay (r = 0.339; $p = 0.002$).

Similarly, patients with a personal history of previous thromboembolic events had lower post-surgery haemoglobin levels ($p = 0.033$) and required fewer units of platelets ($p = 0.039$) and FFP ($p = 0.030$).

Twenty-nine surgeries (32%) occurred in an emergency setting, of which 19 (66%) were abdominal procedures. Patients admitted under these circumstances exhibited significantly lower haemoglobin levels before ($p = 0.003$) and after surgery ($p < 0.001$). They also received more RBC units ($p < 0.001$), FFP ($p = 0.011$), fibrinogen concentrate ($p = 0.019$), and experienced extended hospital stays ($p < 0.001$).

We discovered a tendency toward a positive correlation between BMI and fibrinogen levels (r = 0.326; $p = 0.053$; Figure 5). Additionally, there was a negative correlation between lower post-operative haemoglobin levels and the number of RBC units (r = −0.569; $p < 0.001$), as well as the amount of fibrinogen concentrate administered (r = −0.356; $p = 0.001$), and the length of hospital stay (r = −0.386; $p < 0.001$).

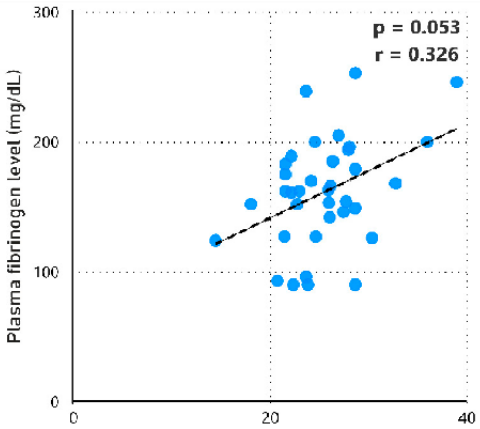

Figure 5. Spearman's correlation of BMI with plasma fibrinogen levels (mg/dL) in all patients. The correlation coefficient, p-value, and best-fit line (dashed line) are displayed. BMI, body mass index.

4. Discussion

Fibrinogen concentrate is widely used in multiple countries to treat acquired hypofibrinogenemia in various clinical scenarios, including surgery [7]. Although many studies have validated its efficacy and safety, there are still conflicting data.

During significant blood loss, fibrinogen levels can decrease rapidly, a condition made worse by hyperfibrinolysis, severe shock, trauma, or acidosis. Moreover, compromised fibrinogen synthesis due to liver disease or hypothermia can contribute to hypofibrinogenemia [3,4].

This prospective observational study concentrated on the safety of administering fibrinogen concentrate in non-trauma and non-obstetric surgical patients since the inherent pathophysiology of coagulopathy could vary from standard surgery. This study primarily considered mortality and thromboembolic events, aiming to encapsulate a real-world approach to bleeding in relation to hypofibrinogenemia.

Despite decades of pharmacovigilance demonstrating a low risk of adverse events in both congenital and acquired fibrinogen deficiencies, concerns persist regarding the potential thromboembolic complications associated with fibrinogen concentrate administration. Adverse events linked to fibrinogen concentrate can include anaphylaxis, hypersensitivity, allergic reactions, thromboembolic complications, and potential virus transmission [7,8]. The latter could also stem from blood component administration or patient behaviour. These are rare occurrences that were not observed in our study.

Our study, which encompassed 91 surgical patients, did not reveal any noticeable associations between the administration of fibrinogen concentrate and mortality among the eight patients who died. Interestingly, six of these patients were admitted in an emergency context with significant comorbidities. Additionally, an association was observed among those who received multiple transfusions and underwent abdominal surgery, although causality remained unclear.

Similarly, eight patients experienced thromboembolic events after surgery, but they were all alive upon discharge from the hospital. Contrary to other reports, there was an equal distribution of sexes among these patients [7,8]. Confounding variables such as concurrent illnesses and the personal or familial history of thrombotic disease could provide alternate explanations for these thromboembolic events. Of these eight patients, only one had no associated comorbidities. Among the remaining seven, one had a positive family history of thrombotic disease. These patients also presented with diverse conditions, including prior thromboembolic events in two cases, a history of COVID-19 infection, cancer, and atrial fibrillation, among others. In this study, a few patients were recruited with conditions that may affect fibrinogen levels. In fact, only one patient had previously known non-congenital hypofibrinogenemia (~90 mg/dL). One patient had a liver transplant 25 years ago but presented with normal fibrinogen levels before surgery. Another patient had chronic liver disease and showed major bleeding after surgery. Regarding known prior haematological malignancies, one patient had myelodysplastic syndrome, one had Non-Hodgkin's Lymphoma, and one had Chronic Lymphatic Leukemic, which also developed an episode of major bleeding after surgery.

Elevated plasma fibrinogen levels have been associated with thrombotic states across various clinical scenarios, such as venous thromboembolism, the rupture of arterial atherosclerotic plaque, and intracardiac thromboembolism [1]. Administering fibrinogen concentrate to achieve haemostasis in patients with major bleeding could disrupt the haemostatic equilibrium and potentially induce a thrombotic state due to its ability to increase plasma fibrinogen levels.

Research from animal and patient trauma models indicates that fibrinogen concentrate may temporarily heighten endogenous thrombin activity at the wound site, stemming from an elevated fibrinogen concentration in the plasma. However, this effect on plasma fibrinogen levels is fleeting, with no observed suppression of endogenous fibrinogen synthesis [25,26].

The transient effect of fibrinogen concentrate could be due to fibrinogen consumption or metabolism. For example, a study involving patients undergoing coronary artery bypass graft surgery showed an increase in D-dimers about 2 h after the administration of fibrinogen concentrate, supporting this idea [27].

Similarly, another study conducted on trauma patients found that plasma fibrinogen levels on day 3, following fibrinogen concentrate administration, resembled those observed in the control group. This observation aligns with the acute-phase response nature of fibrinogen. Its levels fluctuate in response to physiological stressors, reflecting a return to baseline levels over time [27].

The primary objective of our study was not to assess the efficacy of fibrinogen concentrate, which depends on various factors. These include the clinical context, institutional protocols, control of crucial steps, such as the early recognition of significant bleeding, effective team communication, and accepted targets for controlling bleeding coagulopathy, among others [6,15,28].

In terms of safety, which was our study's primary focus, our findings suggest no association between the administration of fibrinogen concentrate in the perioperative setting and an increased risk of thromboembolic events. Of the eight patients who experienced thromboembolic events, each case had other strong factors contributing to these clinical occurrences. This suggests that fibrinogen concentrate administration cannot be solely attributed to these events.

Secondary outcomes align with some conclusions drawn by other authors [29–31]. Age appears to be a risk factor for the necessity of more units of RBC and extended hospital stays. Moreover, patients who endured thromboembolic events during the follow-up phase also had longer hospital stays and received more units of RBC. Our findings indicate that age, the incidence of thromboembolic events, and emergency surgeries are connected to a higher need for blood transfusions and longer hospital stays. Even though the use of fibrinogen concentrate did not seem to heighten the risk of thromboembolic events, patients requiring higher doses of fibrinogen concentrate also received more units of RBC. This finding aligns with a systematic review analysing the use of fibrinogen concentrate for haemorrhages in emergencies [32]. We would also like to underscore the positive correlation between BMI and fibrinogen levels at the deciding point.

We acknowledge several limitations to our study, including the small number of patients recruited, the variety of surgical procedures with differing underlying pathophysiological mechanisms for haemorrhage, and possible confounding variables like emergent situations and comorbidities. We also chose to confine our study protocol to hospital discharge, given the temporary effect of fibrinogen concentrate administration and the potential emergence of other confounding variables during an extended follow-up period in such a varied population.

Nevertheless, our study provides valuable insights into the management of bleeding complications in real-world settings outside the controlled environment of a randomized controlled trial. It underscores the need for special clinical caution in older patients, those undergoing emergency surgeries, the recipients of multiple blood transfusions, and patients with a history of thromboembolic events. Although there were insufficient data for statistical analysis, patients with critically low fibrinogen levels at the time of the decision to administer fibrinogen concentrate may benefit from close monitoring. Indeed, these situations may be analogous to other clinical scenarios where a decrease in fibrinogen is an early predictor of the severity of postpartum haemorrhage [33].

By adhering to institutional bleeding protocols, we ensured the close monitoring of patients throughout our study. Importantly, by focusing on evaluating the safety of fibrinogen concentrate administration in surgical settings rather than its efficacy, we could thoroughly examine adverse events, which is an often-overlooked aspect in other studies.

5. Conclusions

Our results indicate a favourable safety profile for fibrinogen concentrate in surgical patients, demonstrated by a low incidence of deaths and thromboembolic events, which is primarily attributed to other factors. However, future research should prioritize minimizing bias and enhancing statistical robustness to more effectively illuminate clinically significant patient safety measures.

Author Contributions: Conceptualization, M.G. and J.L.; methodology, M.G. and M.Â.-D.; software, M.Â.-D.; validation, M.G. and J.L.; formal analysis, M.Â.-D.; investigation, M.G., M.Â.-D. and J.L.; resources, M.Â.-D.; data curation, M.G. and M.Â.-D.; writing—original draft preparation, M.G. and M.Â.-D.; writing—review and editing, M.G. and J.L.; visualization, M.G. and J.L.; supervision, J.L.; project administration, M.G. and J.L.; funding acquisition, M.G. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: This study was conducted following the Declaration of Helsinki and approved by the Ethics Committee of Hospital da Luz Lisboa (CES/58/2022/ME, approved on 4 July 2022).

Informed Consent Statement: Informed consent was obtained from all subjects involved in this study.

Data Availability Statement: The original contributions presented in the study are included in the article, further inquiries can be directed to the corresponding author.

Acknowledgments: The authors thank Ana Cristina Silva and Pedro Gomes for their contribution to this work, and CSL Behring for the article processing fee of the manuscript.

Conflicts of Interest: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

References

1. Lowe, G.D.; Rumley, A.; Mackie, I.J. Plasma fibrinogen. *Ann. Clin. Biochem.* **2004**, *41*, 430–440. [CrossRef] [PubMed]
2. Besser, M.W.; MacDonald, S.G. Acquired hypofibrinogenemia: Current perspectives. *J. Blood Med.* **2016**, *7*, 217–225. [CrossRef] [PubMed]
3. Sørensen, B.; Larsen, O.H.; Rea, C.J.; Tang, M.; Foley, J.H.; Fenger-Eriksen, C. Fibrinogen as a hemostatic agent. *Semin. Thromb. Hemost.* **2012**, *38*, 268–273. [CrossRef] [PubMed]
4. Levy, J.H.; Szlam, F.; Tanaka, K.A.; Sniecienski, R.M. Fibrinogen and hemostasis: A primary hemostatic target for the management of acquired bleeding. *Anesth. Analg.* **2012**, *114*, 261–274. [CrossRef] [PubMed]
5. Fenger-Eriksen, C.; Ingerslev, J.; Sorensen, B. Fibrinogen concentrate—A potential universal hemostatic agent. *Expert. Opin. Biol. Ther.* **2009**, *9*, 1325–1333. [CrossRef]
6. Kozek-Langenecker, S. Clinical efficacy of fresh frozen plasma compared with coagulation factor concentrates for treating coagulopathy in patients with massive bleeding. *Med. Intensiv.* **2016**, *40*, 371–373. [CrossRef]
7. Solomon, C.; Groner, A.; Ye, J.; Pendrak, I. Safety of fibrinogen concentrate: Analysis of more than 27 years of pharmacovigilance data. *Thromb. Haemost.* **2015**, *113*, 759–771.
8. Costa-Filho, R.; Hochleitner, G.; Wendt, M.; Teruya, A.; Spahn, D.R. Over 50 Years of Fibrinogen Concentrate. *Clin. Appl. Thromb. Hemost.* **2016**, *22*, 109–114. [CrossRef]
9. Kietaibl, S.; Ahmed, A.; Afshari, A.; Albaladejo, P.; Aldecoa, C.; Barauskas, G.; De Robertis, E.; Faraoni, D.; Filipescu, D.C.; Fries, D.; et al. Management of severe peri-operative bleeding: Guidelines from the European Society of Anaesthesiology and Intensive Care: Second update 2022. *Eur. J. Anaesthesiol.* **2023**, *40*, 226–304.
10. Rossaint, R.; Afshari, A.; Bouillon, B.; Cerny, V.; Cimpoesu, D.; Curry, N.; Duranteau, J.; Filipescu, D.; Grottke, O.; Gronlykke, L.; et al. The European guideline on management of major bleeding and coagulopathy following trauma: Sixth edition. *Crit. Care* **2023**, *27*, 80. [CrossRef]
11. Gomes, M.; Angelo-Dias, M.; Duarte, G.S.; Dias, S.S.; Serra, S.S.; Lima, J. Safety of Fibrinogen Concentrate in Non-Trauma and Non-Obstetric Adult Patients during Perioperative Care: Systematic Review and Meta-Analysis. *J. Clin. Med.* **2024**, *13*, 3482. [CrossRef] [PubMed]
12. Fominskiy, E.; Nepomniashchikh, V.A.; Lomivorotov, V.V.; Monaco, F.; Vitiello, C.; Zangrillo, A.; Landoni, G. Efficacy and Safety of Fibrinogen Concentrate in Surgical Patients: A Meta-Analysis of Randomized Controlled Trials. *J. Cardiothorac. Vasc. Anesth.* **2016**, *30*, 1196–1204. [CrossRef] [PubMed]
13. Warmuth, M.; Mad, P.; Wild, C. Systematic review of the efficacy and safety of fibrinogen concentrate substitution in adults. *Acta Anaesthesiol. Scand.* **2012**, *56*, 539–548. [CrossRef] [PubMed]

14. Wikkelso, A.; Lunde, J.; Johansen, M.; Stensballe, J.; Wetterslev, J.; Moller, A.M.; Afshari, A. Fibrinogen concentrate in bleeding patients. *Cochrane Database Syst. Rev.* **2013**, *2013*, CD008864. [CrossRef] [PubMed]
15. Levy, J.H.; Welsby, I.; Goodnough, L.T. Fibrinogen as a therapeutic target for bleeding: A review of critical levels and replacement therapy. *Transfusion* **2014**, *54*, 1389–1405, quiz 1388. [CrossRef]
16. Ranucci, M.; Baryshnikova, E.; Crapelli, G.B.; Rahe-Meyer, N.; Menicanti, L.; Frigiola, A.; Surgical Clinical Outcome REsearch (SCORE) Group. Randomized, double-blinded, placebo-controlled trial of fibrinogen concentrate supplementation after complex cardiac surgery. *J. Am. Heart Assoc.* **2015**, *4*, e002066. [CrossRef]
17. Ng, K.T.; Yap, J.L.L.; Kwok, P.E. The effect of fibrinogen concentrate on postoperative blood loss: A systematic review and meta-analysis of randomized controlled trials. *J. Clin. Anesth.* **2020**, *63*, 109782. [CrossRef]
18. Sabate, A.; Gutierrez, R.; Beltran, J.; Mellado, P.; Blasi, A.; Acosta, F.; Costa, M.; Reyes, R.; Torres, F. Impact of Preemptive Fibrinogen Concentrate on Transfusion Requirements in Liver Transplantation: A Multicenter, Randomized, Double-Blind, Placebo-Controlled Trial. *Am. J. Transplant.* **2016**, *16*, 2421–2429. [CrossRef]
19. Roy, A.; Stanford, S.; Nunn, S.; Alves, S.; Sargant, N.; Rangarajan, S.; Smith, E.A.; Bell, J.; Dayal, S.; Cecil, T.; et al. Efficacy of fibrinogen concentrate in major abdominal surgery—A prospective, randomized, controlled study in cytoreductive surgery for pseudomyxoma peritonei. *J. Thromb. Haemost.* **2020**, *18*, 352–363. [CrossRef]
20. Callum, J.; Farkouh, M.E.; Scales, D.C.; Heddle, N.M.; Crowther, M.; Rao, V.; Hucke, H.P.; Carroll, J.; Grewal, D.; Brar, S.; et al. Effect of Fibrinogen Concentrate vs Cryoprecipitate on Blood Component Transfusion After Cardiac Surgery: The FIBRES Randomized Clinical Trial. *JAMA* **2019**, *322*, 1966–1976. [CrossRef]
21. Rahe-Meyer, N.; Levy, J.H.; Mazer, C.D.; Schramko, A.; Klein, A.A.; Brat, R.; Okita, Y.; Ueda, Y.; Schmidt, D.S.; Ranganath, R.; et al. Randomized evaluation of fibrinogen vs placebo in complex cardiovascular surgery (REPLACE): A double-blind phase III study of haemostatic therapy. *Br. J. Anaesth.* **2016**, *117*, 41–51. [CrossRef] [PubMed]
22. Rahe-Meyer, N.; Solomon, C.; Hanke, A.; Schmidt, D.S.; Knoerzer, D.; Hochleitner, G.; Sorensen, B.; Hagl, C.; Pichlmaier, M. Effects of fibrinogen concentrate as first-line therapy during major aortic replacement surgery: A randomized, placebo-controlled trial. *Anesthesiology* **2013**, *118*, 40–50. [CrossRef] [PubMed]
23. Najafi, A.; Shariat Moharari, R.; Orandi, A.A.; Etezadi, F.; Sanatkar, M.; Khajavi, M.R.; Ahmadi, A.; Pourfakhr, P.; Imani, F.; Mojtahedzadeh, M.; et al. Prophylactic administration of fibrinogen concentrate in perioperative period of total hip arthroplasty: A randomized clinical trial study. *Acta Med. Iran.* **2014**, *52*, 804–810.
24. Direção Geral da Saúde. Abordagem da Transfusão Maciça no Adulto. Recommendation nr. 011/2013, Updated on 18 July 2017. Available online: https://normas.dgs.min-saude.pt/2013/07/30/abordagem-da-transfusao-macica-no-adulto/ (accessed on 5 August 2024).
25. Zentai, C.; Solomon, C.; van der Meijden, P.E.; Spronk, H.M.; Schnabel, J.; Rossaint, R.; Grottke, O. Effects of Fibrinogen Concentrate on Thrombin Generation, Thromboelastometry Parameters, and Laboratory Coagulation Testing in a 24-Hour Porcine Trauma Model. *Clin. Appl. Thromb. Hemost* **2016**, *22*, 749–759. [CrossRef] [PubMed]
26. Zentai, C.; Braunschweig, T.; Schnabel, J.; Rose, M.; Rossaint, R.; Grottke, O. Fibrinogen concentrate does not suppress endogenous fibrinogen synthesis in a 24-hour porcine trauma model. *Anesthesiology* **2014**, *121*, 753–764. [CrossRef]
27. Schlimp, C.J.; Ponschab, M.; Voelckel, W.; Treichl, B.; Maegele, M.; Schochl, H. Fibrinogen levels in trauma patients during the first seven days after fibrinogen concentrate therapy: A retrospective study. *Scand. J. Trauma. Resusc. Emerg. Med.* **2016**, *24*, 29. [CrossRef]
28. Carvalho, M.; Rodrigues, A.; Gomes, M.; Carrilho, A.; Nunes, A.R.; Orfao, R.; Alves, A.; Aguiar, J.; Campos, M. Interventional Algorithms for the Control of Coagulopathic Bleeding in Surgical, Trauma, and Postpartum Settings: Recommendations From the Share Network Group. *Clin. Appl. Thromb. Hemost.* **2016**, *22*, 121–137. [CrossRef]
29. Buhl, H.; Norgaard, A.; Otkjaer, A.; Jorgensen, L.N.; Jorgensen, H.L. The impact of anemia and blood transfusion on mortality after open abdominal surgery in the elderly. *Langenbecks Arch. Surg.* **2023**, *408*, 421. [CrossRef]
30. Ming, Y.; Liu, J.; Zhang, F.; Chen, C.; Zhou, L.; Du, L.; Yan, M. Transfusion of Red Blood Cells, Fresh Frozen Plasma, or Platelets Is Associated With Mortality and Infection After Cardiac Surgery in a Dose-Dependent Manner. *Anesth. Analg.* **2020**, *130*, e32. [CrossRef]
31. Sihler, K.C.; Napolitano, L.M. Complications of massive transfusion. *Chest* **2010**, *137*, 209–220. [CrossRef]
32. Itagaki, Y.; Hayakawa, M.; Takahashi, Y.; Hirano, S.; Yamakawa, K. Emergency administration of fibrinogen concentrate for haemorrhage: Systematic review and meta-analysis. *World J. Emerg. Surg.* **2023**, *18*, 27. [CrossRef] [PubMed]
33. Charbit, B.; Mandelbrot, L.; Samain, E.; Baron, G.; Haddaoui, B.; Keita, H.; Sibony, O.; Mahieu-Caputo, D.; Hurtaud-Roux, M.F.; Huisse, M.G.; et al. sThe decrease of fibrinogen is an early predictor of the severity of postpartum hemorrhage. *J. Thromb. Haemost.* **2007**, *5*, 266–273. [CrossRef] [PubMed]

CHAPTER V

General Discussion

There is a fine line between thrombosis and hemostasis. In real-world clinical settings, away from the controlled environment of randomized trials, patients with bleeding often face resource disparities and decisions made under high-pressure circumstances. These realities frequently obscure the outcomes of therapies aimed at controlling hemostasis. In the field of surgery, minor hemorrhages, usually manageable and requiring no specific intervention, can become serious issues when coagulopathy develops and is untreated promptly or appropriately. Similarly, surgical hemorrhages that are initially manageable can escalate into life-threatening situations if overlooked.

During significant blood loss, fibrinogen levels can plummet rapidly, further aggravated by factors like hyperfibrinolysis, trauma, severe shock, or acidosis. Liver disease and hypothermia can additionally hinder fibrinogen synthesis, contributing to hypofibrinogenemia [46]. Fibrinogen concentrate is extensively used in numerous countries to treat acquired hypofibrinogenemia, especially in surgical settings [12, 27]. This practice adheres to guidelines issued by the European Society of Anaesthesiology and Intensive Care, which have endorsed its use for perioperative bleeding [36]. Regardless of substantial research and decades of pharmacovigilance, inconsistent data concerning the efficacy and safety of fibrinogen concentrate continue to exist.

Originally, fibrinogen concentrate was utilized for the treatment of congenital fibrinogen deficiencies such as hypofibrinogenemia, afibrinogenemia, and dysfibrinogenemia. These conditions demanded smaller therapeutic doses and were applied to both pediatric and adult patients. Over the past three decades, the therapeutic spectrum of fibrinogen concentrate has widened to encompass acquired conditions such as perioperative bleeding, post-partum hemorrhage, trauma-related coagulopathy, and gastrointestinal bleeding [27, 28] [47, 48]. While fibrinogen supplementation aims to achieve hemostasis in bleeding patients, heightened fibrinogen levels have been associated with thrombotic states in various clinical contexts, including venous thromboembolism, rupture of arterial plaques, and intracardiac thromboembolism [10]. This dual role underscores the potential risks of fibrinogen concentrate in disrupting hemostatic equilibrium and inducing thrombotic states.

Research, conducted using both animal trauma models and clinical data, demonstrates that fibrinogen concentrate can, temporarily, increase thrombin activity at wound locations due to elevated plasma fibrinogen levels. However, this effect is short-lived and does not inhibit endogenous fibrinogen production [49, 50]. Human studies have shown that plasma fibrinogen levels return to normal within days of administration, aligning with the acute-phase response. One particular study found that trauma patients had fibrinogen levels comparable to those of control groups by the third day post-administration [51]. Likewise, studies involving patients undergoing coronary artery bypass grafting noted brief increases in D-dimer levels following fibrinogen concentrate administration, which is indicative of fibrinogen metabolism and consumption [51].

A systematic review was conducted to consolidate and understand the safety of fibrinogen concentrate, specifically focusing on non-trauma, non-obstetric adult surgical patients. This review incorporated ten studies that included 1,391 patients and produced significant findings. It was discovered that fibrinogen concentrate was connected to an overall risk reduction of thromboembolic events in perioperative settings. Although cardiovascular surgery studies dominated the review, there were no significant differences in thromboembolic events between the fibrinogen concentrate group and the control group. Mortality data were limited; in the studies where mortality was addressed, it was not directly associated with the use of fibrinogen concentrate. However, drawing definitive conclusions was challenging due to heterogeneity among the studies, stemming from variations in protocols, thresholds, and dosages.

To the best of our knowledge, no published systematic reviews have reported findings that align with this study, distinguishing it from prior research. Several limitations in previous reviews are worth acknowledgment. For example, Fominskiy and Wikkelso's [31, 52] meta-analyses encompassed both pediatric and adult populations. Such an approach might bias the findings since fibrinogen levels significantly vary with age [53]. Secondly, the inclusion of a post-partum hemorrhage study by Fominskiy et al. [52] adds an additional layer of heterogeneity, considering the physiological variations in fibrinogen levels associated with pregnancy. During pregnancy, fibrinogen levels typically increase and then sharply decrease after delivery. In post-partum hemorrhage scenarios, fibrinogen levels below 200 mg/dl strongly indicate severe hemorrhage [54]. Moreover, the WOMAN trial revealed that hypofibrinolytic conditions in post-partum hemorrhage, effectively treated with tranexamic acid, involve unique pathophysiological mechanisms compared to surgical bleeding [55]. Although these mechanisms sometimes also require critical control, they are different from those in surgical situations.

Trauma patients were also included in these meta-analyses, which could skew mortality outcomes due to the unique factors influencing bleeding in trauma situations, such as tissue injury and shock. These conditions often lead to a specific type of acquired coagulopathy characterized by dilution, acidosis, hyperfibrinolysis, extremely low fibrinogen levels, and activation of the protein C pathway, which differs significantly from surgical coagulopathies [56, 57]. Furthermore, while the meta-analysis performed by Ka Ting et al. [58] aimed to reduce heterogeneity, its reliance on a limited number of databases might have excluded relevant studies, potentially affecting the comprehensiveness of its findings. Finally, other systematic reviews with meta-analyses either include a limited number of studies or focus solely on specific clinical settings, surgeries, or comparators. This narrow scope impedes their ability to provide a broad evaluation of fibrinogen concentrate effectiveness and safety in managing bleeding [32, 59, 60].

Building on the conclusions of the systematic review and meta-analysis, a single-center observational study was carried out to assess the safety of fibrinogen concentrate in non-trauma,

non-obstetric surgical patients. The goal of this study was to offer a real-world perspective on managing bleeding associated with hypofibrinogenemia, based on the analysis of data from 91 patients. The primary outcomes were mortality and thromboembolic events.

Eight patients experienced thromboembolic events, yet survived until discharge. Notably, in contrast to other reports, there was no gender predominance among these patients [41]. Seven of those individuals had significant comorbidities such as cancer, atrial fibrillation, historical thromboembolic events, or a history of COVID-19 infection, all of which may have played a role in their outcomes. The overall mortality rate stood at 8.8%, with fatalities primarily resulting from septic, cardiogenic, or hypovolemic shock leading to multi-organ failure. Recognized risk factors for heightened mortality and thromboembolic events included emergency operations, advanced age, depleted pre-administration fibrinogen levels, and the necessity for multiple blood transfusions. These findings are in agreement with a systematic review concerning the use of fibrinogen concentrate in emergency environments, which confirmed its safety [60].

No significant associations were observed between the administration of fibrinogen concentrate and mortality or adverse events in this study. While the use of fibrinogen concentrate did not seem to raise the risk of thromboembolic events, patients who needed higher doses of fibrinogen concentrate typically received more units of red blood cells. This finding aligns with previous systematic reviews analyzing the use of fibrinogen concentrate in emergency hemorrhage scenarios [60]. Notably, the doses of fibrinogen concentrate were comparable in terms of mortality or thromboembolic events, suggesting these outcomes were not directly associated with the administration of fibrinogen concentrate. Intriguingly, abdominal surgeries were linked with the majority of the fatalities, contrasting with just one death following cardiac surgery, although the reasons for this discrepancy remain unclear.

In conclusion, this observational study offered valuable insights into the practical application of fibrinogen concentrate, thus supplementing the conclusions of the systematic review and meta-analysis. Unlike the carefully regulated environment of randomized clinical trials, this research emphasized real-world challenges and reinforced the safety profile of fibrinogen concentrate, furthering our comprehension of its implementation in a variety of clinical situations.

Strengths and limitations

This study possesses several noteworthy strengths. Unlike other investigations on this subject, our systematic review concentrated solely on a specific patient group, reducing the possible bias and heterogeneity that frequently occur due to the inclusion of diverse population characteristics. However, none of the included studies were expressly designed to evaluate safety as the primary outcome. The review's findings are bolstered by the accompanying prospective study, offering valuable insights into handling bleeding complications in real-world clinical

scenarios, beyond the controlled environment of randomized controlled trials. Key considerations arose from the collected data regarding certain hospital populations, particularly those undergoing either scheduled or urgent/emergency surgeries, bearing significant practical implications.

This study highlights the necessity for increased clinical vigilance for older patients, individuals undergoing emergency surgeries, recipients of multiple blood transfusions, and those with a history of thromboembolic events. While the statistical analysis was impeded by inadequate data, it is clear that patients displaying critically low fibrinogen levels at the time of fibrinogen concentrate administration require careful monitoring. This mirrors other clinical conditions where a decrease in fibrinogen acts as an early indicator of severity, such as in the case of PPH.

While these studies provide valuable insights, there are some acknowledged limitations. Many of the included studies had small sample sizes, which could potentially limit their ability to detect rare adverse events. Despite identifying a significant number of studies reporting safety outcomes following the administration of fibrinogen concentrate, considerable heterogeneity persists. Variations in the countries where the studies were conducted, the dominance of data from cardiac surgery, differences in administration methods, thresholds for fibrinogen levels or point-of-care test targets, and the range of fibrinogen concentrate dosages across trials all complicate the generalization and interpretation of the findings. Furthermore, the relevance of older studies might be questioned due to the evolution of care standards and advancements in blood management protocols.

The heterogeneity and limited number of studies create uncertainties about the applicability of our findings to other types of surgeries, such as abdominal, genitourinary, or cancer-related procedures. Nonetheless, our analysis did not present evidence of publication bias. This suggests that the observed reduction in thromboembolic events in the fibrinogen concentrate arm was not unduly swayed by selectively reported positive outcomes.

Assessing mortality is a vital aspect of evaluating the safety of any intervention. Although the inclusion of mortality as a primary outcome might be controversial, it is crucial to recognize the inherent complexity in attributing death solely to an intervention. This process fails to account for other contributing factors like patient comorbidities, surgical risk, age, and intra- and postoperative complications. Unfortunately, several studies do not adequately determine whether the mortality events were definitely or likely related to the intervention. This situation calls for caution when interpreting these results.

The observational study had some limitations as well, such as a small sample size and the inclusion of various surgical procedures with different underlying mechanisms of hemorrhage. Factors confounding the results, including emergent situations and patient comorbidities, further complicated the study's findings. Moreover, our study protocol was restricted to the period of hospital discharge. This restriction was due to the transient effects of fibrinogen

concentrate and the possibility that other confounding variables could emerge during a prolonged follow-up in such a diversified population.

Conclusions and future perspectives

The results from both the systematic review and the prospective study are closely aligned, reinforcing similar conclusions. The systematic review suggests that the use of fibrinogen concentrate in the perioperative care of non-trauma, non-obstetric adult patients may provide significant benefits. Complementing this, the prospective study sheds light on a favorable safety profile for fibrinogen concentrate, with a low incidence of deaths and thromboembolic events that are largely attributable to other factors. However, future research should aim to reduce bias and improve statistical rigor to better elucidate clinically relevant safety outcomes.

Adherence to institutional bleeding protocols played a pivotal role in ensuring comprehensive patient monitoring throughout our study and is highly recommended for other institutions. This study zeroed in on the safety implications of fibrinogen concentrate administration in surgical settings rather than its efficacy, thereby providing a detailed evaluation of adverse events – an aspect that existing literature often underemphasizes.

Alisa Wolberg aptly stated that fibrinogen is an extraordinary molecule with multifaceted functions. This statement equally applies to its insoluble form, fibrin. Both fibrinogen and fibrin are implicated in numerous diseases. Studying the structure and function of fibrin clots in greater detail can provide valuable insights into disease risks. When we bridge basic science with clinical investigation in this field, it opens up new horizons. For instance, if clot structure is critical for stability, how can we develop standardized, *in vivo* methods to study it?

The role of fibrinogen isoforms also represents a promising avenue for research. Quantifying these isoforms in bleeding patients could provide essential information about the appropriate dose and anticipated response to fibrinogen administration, tailored to specific clinical scenarios. This precision-based approach may optimize patient outcomes and reduce variability in treatment effectiveness.

The importance of educating and updating healthcare teams on the mechanisms of hemostasis and the significance of bleeding control cannot be overstated. Though often deemed complex or monotonous, an in-depth understanding of hemostasis can enhance patient outcomes and even save lives. Moreover, it addresses an additional urgent issue: the conservation of allogeneic blood, a resource that is becoming increasingly scarce due to a variety of factors. On the other hand, and perhaps more importantly now, each institution should focus on educating and updating their teams on how hemostasis works and the importance of bleeding control. Although the mechanisms of hemostasis are often perceived as complex and tedious, a thorough understanding can significantly improve patient outcomes and save lives. Moreover, it serves

another vital purpose: conserving allogeneic blood, which is becoming increasingly difficult to source in today's world for various reasons.

Finally, but not less importantly, the standardization of procedures through institutional protocols for bleeding patients is of paramount importance. This involves assembling well-trained teams that are equipped with advanced diagnostic tools for assessing hemostasis. They should also have clear guidelines for administering the appropriate hemostatic agents, such as fibrinogen concentrate, in the correct dose at the proper time. This comprehensive approach can bolster patient care and improve resource management, ensuring optimal outcomes in critical bleeding scenarios.

REFERENCES

Note: **additional references for each study were placed in their respective chapter**

1. Al-Koussa, H., I. AlZaim, and M.E. El-Sabban, *Pathophysiology of Coagulation and Emerging Roles for Extracellular Vesicles in Coagulation Cascades and Disorders.* J Clin Med, 2022. **11**(16).
2. Hoffman, M., *A cell-based model of coagulation and the role of factor VIIa.* Blood Rev, 2003. **17 Suppl 1**: p. S1-5.
3. Gomes, M., *Plasma alternatives in acquired bleeding disorders — factor concentrates.* Journal of Transfusion Medicine, 2014. **7**(1): p. 15-19.
4. Smith, S.A., *The cell-based model of coagulation.* J Vet Emerg Crit Care (San Antonio), 2009. **19**(1): p. 3-10.
5. May, J.E., A.S. Wolberg, and M.Y. Lim, *Disorders of Fibrinogen and Fibrinolysis.* Hematol Oncol Clin North Am, 2021. **35**(6): p. 1197-1217.
6. Wolberg, A.S., *Fibrinogen and fibrin: synthesis, structure, and function in health and disease.* J Thromb Haemost, 2023. **21**(11): p. 3005-3015.
7. Chapin, J.C. and K.A. Hajjar, *Fibrinolysis and the control of blood coagulation.* Blood Rev, 2015. **29**(1): p. 17-24.
8. Gue, Y.X., et al., *Assessment of endogenous fibrinolysis in clinical practice using novel tests: ready for clinical roll-out?* SN Applied Sciences, 2021. **3**(5): p. 524.
9. Levi, M., et al., *Infection and inflammation and the coagulation system.* Cardiovasc Res, 2003. **60**(1): p. 26-39.
10. Lowe, G.D., A. Rumley, and I.J. Mackie, *Plasma fibrinogen.* Ann Clin Biochem, 2004. **41**(Pt 6): p. 430-40.
11. Kamath, S. and G.Y. Lip, *Fibrinogen: biochemistry, epidemiology and determinants.* QJM, 2003. **96**(10): p. 711-29.
12. Besser, M.W. and S.G. MacDonald, *Acquired hypofibrinogenemia: current perspectives.* J Blood Med, 2016. **7**: p. 217-225.
13. Ternstrom, L., et al., *Plasma activity of individual coagulation factors, hemodilution and blood loss after cardiac surgery: a prospective observational study.* Thromb Res, 2010. **126**(2): p. e128-33.
14. Solomon, C., C. Hagl, and N. Rahe-Meyer, *Time course of haemostatic effects of fibrinogen concentrate administration in aortic surgery.* Br J Anaesth, 2013. **110**(6): p. 947-56.
15. Hiippala, S.T., G.J. Myllyla, and E.M. Vahtera, *Hemostatic factors and replacement of major blood loss with plasma-poor red cell concentrates.* Anesth Analg, 1995. **81**(2): p. 360-5.
16. Schochl, H., M. Maegele, and W. Voelckel, *Fixed ratio versus goal-directed therapy in trauma.* Curr Opin Anaesthesiol, 2016. **29**(2): p. 234-44.
17. Gorlinger, K., et al., *The role of evidence-based algorithms for rotational thromboelastometry-guided bleeding management.* Korean J Anesthesiol, 2019. **72**(4): p. 297-322.
18. Gorlinger, K., et al., *Reduction of Fresh Frozen Plasma Requirements by Perioperative Point-of-Care Coagulation Management with Early Calculated Goal-Directed Therapy.* Transfus Med Hemother, 2012. **39**(2): p. 104-113.
19. Whiting, D. and J.A. DiNardo, *TEG and ROTEM: technology and clinical applications.* Am J Hematol, 2014. **89**(2): p. 228-32.
20. Leal-Noval, S.R., et al., *Administration of fibrinogen concentrate for refractory bleeding in massively transfused, non-trauma patients with coagulopathy: a retrospective study with comparator group.* BMC Anesthesiol, 2014. **14**: p. 109.
21. Williams, B., et al., *Practical Use of Thromboelastometry in the Management of Perioperative Coagulopathy and Bleeding.* Transfus Med Rev, 2017. **31**(1): p. 11-25.

22. Carvalho, M., et al., *Interventional Algorithms for the Control of Coagulopathic Bleeding in Surgical, Trauma, and Postpartum Settings: Recommendations From the Share Network Group.* Clin Appl Thromb Hemost, 2016. **22**(2): p. 121-37.
23. Gomes, M., et al., *Safety of Fibrinogen Concentrate in Non-Trauma and Non-Obstetric Adult Patients during Perioperative Care: Systematic Review and Meta-Analysis.* J Clin Med, 2024. **13**(12).
24. Wolberg, A.S. and Y. Sang, *Fibrinogen and Factor XIII in Venous Thrombosis and Thrombus Stability.* Arterioscler Thromb Vasc Biol, 2022. **42**(8): p. 931-941.
25. Levy, J.H., et al., *Therapeutic Plasma Transfusion in Bleeding Patients: A Systematic Review.* Anesth Analg, 2017. **124**(4): p. 1268-1276.
26. Kozek-Langenecker, S., *Clinical efficacy of fresh frozen plasma compared with coagulation factor concentrates for treating coagulopathy in patients with massive bleeding.* Med Intensiva, 2016. **40**(6): p. 371-3.
27. Levy, J.H., et al., *Fibrinogen and hemostasis: a primary hemostatic target for the management of acquired bleeding.* Anesth Analg, 2012. **114**(2): p. 261-74.
28. Fenger-Eriksen, C., J. Ingerslev, and B. Sorensen, *Fibrinogen concentrate--a potential universal hemostatic agent.* Expert Opin Biol Ther, 2009. **9**(10): p. 1325-33.
29. Nascimento, B., et al., *Cryoprecipitate transfusion in bleeding patients.* CJEM, 2020. **22**(S2): p. S4-S11.
30. Costa-Filho, R., et al., *Over 50 Years of Fibrinogen Concentrate.* Clin Appl Thromb Hemost, 2016. **22**(2): p. 109-14.
31. Wikkelso, A., et al., *Fibrinogen concentrate in bleeding patients.* Cochrane Database Syst Rev, 2013. **2013**(8): p. CD008864.
32. Stabler, S.N., et al., *Use of fibrinogen concentrate for trauma-related bleeding: A systematic-review and meta-analysis.* J Trauma Acute Care Surg, 2020. **89**(6): p. 1212-1224.
33. Hofer, S., et al., *Haemostatic support in postpartum haemorrhage: A review of the literature and expert opinion.* Eur J Anaesthesiol, 2023. **40**(1): p. 29-38.
34. Fenger-Eriksen, C., et al., *Fibrinogen substitution improves whole blood clot firmness after dilution with hydroxyethyl starch in bleeding patients undergoing radical cystectomy: a randomized, placebo-controlled clinical trial.* J Thromb Haemost, 2009. **7**(5): p. 795-802.
35. Gomes, M., et al., *Portuguese Consensus and Recommendations for Acquired Coagulopathic Bleeding Management (CCBM).* Clin Appl Thromb Hemost, 2021. **27**: p. 10760296211003984.
36. Kietaibl, S., et al., *Management of severe peri-operative bleeding: Guidelines from the European Society of Anaesthesiology and Intensive Care: Second update 2022.* Eur J Anaesthesiol, 2023. **40**(4): p. 226-304.
37. Tanaka, K.A., et al., *Transfusion and hematologic variables after fibrinogen or platelet transfusion in valve replacement surgery: preliminary data of purified lyophilized human fibrinogen concentrate versus conventional transfusion.* Transfusion, 2014. **54**(1): p. 109-18.
38. Roy, A., et al., *Efficacy of fibrinogen concentrate in major abdominal surgery - A prospective, randomized, controlled study in cytoreductive surgery for pseudomyxoma peritonei.* J Thromb Haemost, 2020. **18**(2): p. 352-363.
39. Rahe-Meyer, N., et al., *Effects of fibrinogen concentrate as first-line therapy during major aortic replacement surgery: a randomized, placebo-controlled trial.* Anesthesiology, 2013. **118**(1): p. 40-50.
40. Bilecen, S., et al., *Effect of Fibrinogen Concentrate on Intraoperative Blood Loss Among Patients With Intraoperative Bleeding During High-Risk Cardiac Surgery: A Randomized Clinical Trial.* JAMA, 2017. **317**(7): p. 738-747.

41. Solomon, C., et al., *Safety of fibrinogen concentrate: analysis of more than 27 years of pharmacovigilance data.* Thromb Haemost, 2015. **113**(4): p. 759-71.
42. Sanders, S., et al., *Fibrinogen Concentrate in the Special Operations Forces Environment.* Mil Med, 2018. **183**(1-2): p. e45-e50.
43. Page, M.J., J.P.T. Higgins, and J.A.C. Sterne, *Chapter 13: Assessing risk of bias due to missing results in a synthesis. In: Higgins JPT, Thomas J, Chandler J, Cumpston M, Li T, Page MJ and Welch VA. Cochrane Handbook for Systematic Reviews of Interventions version 6.4 (updated August 2023). Cochrane, 2023. https://www.training.cochrane.org/handbook/current/chapter-13.*
44. Peryer, G., et al., *Chapter 19: Adverse effects. In: Higgins JPT, Thomas J, Chandler J, Cumpston M, Li T, Page MJ, and Welch VA. Cochrane Handbook for Systematic Reviews of Interventions version 6.4 (updated August 2023). Cochrane, 2023. https://www.training.cochrane.org/handbook/current/chapter-09.*
45. Rahe-Meyer, N., et al., *Randomized evaluation of fibrinogen vs placebo in complex cardiovascular surgery (REPLACE): a double-blind phase III study of haemostatic therapy.* Br J Anaesth, 2016. **117**(1): p. 41-51.
46. Curnow, J., L. Pasalic, and E.J. Favaloro, *Why Do Patients Bleed?* Surg J (N Y), 2016. **2**(1): p. e29-e43.
47. Stanford, S., et al., *Differences in coagulation-relevant parameters: Comparing cryoprecipitate and a human fibrinogen concentrate.* PLoS One, 2023. **18**(8): p. e0290571.
48. Rodrigues, A., et al., *Interventional Algorithm in Gastrointestinal Bleeding-An Expert Consensus Multimodal Approach Based on a Multidisciplinary Team.* Clin Appl Thromb Hemost, 2020. **26**: p. 1076029620931943.
49. Zentai, C., et al., *Effects of Fibrinogen Concentrate on Thrombin Generation, Thromboelastometry Parameters, and Laboratory Coagulation Testing in a 24-Hour Porcine Trauma Model.* Clin Appl Thromb Hemost, 2016. **22**(8): p. 749-759.
50. Zentai, C., et al., *Fibrinogen concentrate does not suppress endogenous fibrinogen synthesis in a 24-hour porcine trauma model.* Anesthesiology, 2014. **121**(4): p. 753-64.
51. Schlimp, C.J., et al., *Fibrinogen levels in trauma patients during the first seven days after fibrinogen concentrate therapy: a retrospective study.* Scand J Trauma Resusc Emerg Med, 2016. **24**: p. 29.
52. Fominskiy, E., et al., *Efficacy and Safety of Fibrinogen Concentrate in Surgical Patients: A Meta-Analysis of Randomized Controlled Trials.* J Cardiothorac Vasc Anesth, 2016. **30**(5): p. 1196-204.
53. Appel, I.M., et al., *Age dependency of coagulation parameters during childhood and puberty.* J Thromb Haemost, 2012. **10**(11): p. 2254-63.
54. Charbit, B., et al., *The decrease of fibrinogen is an early predictor of the severity of postpartum hemorrhage.* J Thromb Haemost, 2007. **5**(2): p. 266-73.
55. Collaborators, W.T., *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.* Lancet, 2017. **389**(10084): p. 2105-2116.
56. Rossaint, R., et al., *The European guideline on management of major bleeding and coagulopathy following trauma: sixth edition.* Crit Care, 2023. **27**(1): p. 80.
57. Davenport, R., *Pathogenesis of acute traumatic coagulopathy.* Transfusion, 2013. **53 Suppl 1**: p. 23S-27S.
58. Ng, K.T., J.L.L. Yap, and P.E. Kwok, *The effect of fibrinogen concentrate on postoperative blood loss: A systematic review and meta-analysis of randomized controlled trials.* J Clin Anesth, 2020. **63**: p. 109782.

59. Warmuth, M., P. Mad, and C. Wild, *Systematic review of the efficacy and safety of fibrinogen concentrate substitution in adults.* Acta Anaesthesiol Scand, 2012. **56**(5): p. 539-48.
60. Itagaki, Y., et al., *Emergency administration of fibrinogen concentrate for haemorrhage: systematic review and meta-analysis.* World J Emerg Surg, 2023. **18**(1): p. 27.