RECOMBINANT ACTIVATED FACTOR VII (rFVIIa/ NovoSeven®) IN THE MANAGEMENT OF MASSIVE BLEEDING IN HOSPITAL UNIVERSITI SAINS MALAYSIA

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2017

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by

NURFATIN MOHD SHAH

Thesis submitted in partial fulfilment of the requirement for the Degree of Master of Science (Transfusion Science)

September 2017

ACKNOWLEDGEMENT

In the name of ALLAH, the most gracious and the most merciful;

First and foremost, I would like to express my gratitude to Allah S.W.T for the blessing to complete this thesis. He is ultimately we depend on for sustenance and guidance.

Thanks for everyone who helped me in completing this work. I submit my highest appreciation to my supervisor, Dr. Chong Soon Eu, who helped in every step of the way with relentless support and motivation at all times. He inspired me greatly to work in this project. I also would like to express my thanks to my field supervisor, Dr. Mohd Zulfakar Mazlan and co-supervisor, Dr. Siti Mardhiana Mohamad who gave full support for this research.

My deepest appreciation to my mother, Sopiah Ahmad for unfailing support and continuous encouragement throughout my study. I am particularly grateful for the assistance given by Nur Sakinah Soid, Nabilah Hanim and all my classmates. Lastly, I would like to thank everyone who has contributed directly or indirectly to this research. This accomplishment would have not been possible without them.

Thank you.

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LIST OF ABBREVIATIONS

rFVIIa	Recombinant Activated Factor VII
kDa	kilo Dalton
pH	Potential of hydrogen
µ	micro
MW	Molecular weight
˚C	Degree celcius
mL	millilitre
mg	milligram
DNA	Deoxyribonucleic acid

ABSTRAK

Pendarahan yang teruk adalah salah satu faktor penyumbang kematian di dunia. Rawatan dalam keadaan pendarahan teruk adalah penting bagi mengembalikan hemostasis pesakit. Beberapa intervensi pernah digunakan untuk menghentikan perdarahan dengan kadar keberkesanan, keselamatan dan keputusan yang berbeza. Salah satu daripada agen hemostasis novel ialah *recombinant activated factor VII* (rFVIIa). Pelbagai kontroversi terhadap penggunaan rFVIIa sebagai salah satu pilihan untuk untuk rawatan pendarahan dalam kalangan pesakit bukan hemofilia. Tujuan kajian ini dijalankan adalah untuk menyelidik penggunaan rFVIIa secara luar indikasi dalam kalangan pesakit yang menagalami pendarahan yang **t**eruk di Hospital Universiti Sains Malaysia.

Rekod perubatan 76 pesakit bukan penghidap hemofilia yang dirawat dengan menggunakan rFVIIa untuk pendarahan, sepanjang tempoh 11 tahun di institusi tunggal yang menggunakan rFVIIa dalam rawatan dikaji untuk indikasi rawatan, kematian dalam masa 24 jam dan 30 hari, keperluan transfusi dan profil pembekuan. Pesakit dikelaskan kepada trauma, selepas bersalin, lain-lain pembedahan dan masalah kesihatan lain. Risiko masalah *thromboembolism* dinilai dalam kalangan pesakit.

Pesakit masalah kesihatan lain adalah yang paling banyak menggunakan rFVIIa diikuti oleh trauma, lain-lain pembedahan, dan paling sedikit diberikan kepada pesakit selepas bersalin. Secara keseluruhan, setiap kategori mencatatkan kurang daripada 25% risiko kematian dalam masa 24 jam manakala pelbagai peratusan untuk kematian pada hari ke-30. Komponen dan produk darah (sel kejap, plasma beku segar, platelet, *cryoprecipitate*) yang diperlukan adalah berkurangan setelah pesakit

menerima rFVIIa. Terdapat juga perubahan profil pembekuan (prothrombin time, activated partial thromboplastin time, international normalized ratio) pada pesakit sebelum dan selepas dirawat dengan rFVIIa. Tiada komplikasi *thromboembolism* dicatatkan dalam kajian ini. Kepentingan statistik telah dinilai pada $p < 0.05$.

Penggunaan rFVIIa adalah efisien dalam memberi kesan yang cepat untuk merawat pendarahan yang teruk. Pesakit yang dirawat dengan rFVIIa menunjukkan pengurangan keperluan transfusi komponen dan produk darah. Selain itu, penambaikan yang ketara dalam parameter pembekuan juga dapat diperhati dalam kalangan pesakit bukan hemophilia di Hospital Universiti Sains Malaysia. Kajian ini mendapati rFVIIa tidak mempunyai kaitan dengan komplikasi *thromboembolism* selepas pembedahan.

ABSTRACT

Massive bleeding has been a one of the contributing causes of death in the world. It is crucial for physicians to establish haemostasis during this condition. Currently, there are various type of methods to arrest bleeding with different efficiency, safety profile and outcome. One of the novel haemostatic agent is recombinant activated factor VII (rFVIIa). There has been a considerable controversy on usage of rFVIIa as one of the option to stop bleeding among non-haemophiliac patients. The aim of this study is to determine the outcome of off-label use of rFVIIa to treat bleeding among the non-haemophiliac patients in Hospital Universiti Sains Malaysia.

Medical records of 76 non-haemophiliac patients treated with rFVIIa for desperate bleeding over 11-year period at the single institution administering rFVIIa were recorded for treatment indications, 24-hours and 30-day mortality, transfusion need and coagulation profiles. The patients were classified into trauma, post-partum, other surgery and medical. Complication of thromboembolism events are assessed among patients.

rFVIIa are most administered in medical patients followed by trauma, other surgery and post-partum. Overall, each category reported less than 25% risk of 24-hour mortality while various percentage for mortality at day-30. Blood and blood products (packed red blood cell, fresh frozen plasma, platelet, cryoprecipitate) needed are significantly reduced in patients receiving rFVIIa. There is also improvement of coagulation profiles (prothrombin time, activated partial thromboplastin time, international normalized ratio) in patients before and after treated with rFVIIa. None

of thromboembolism events described for this study. Statistical significance was assessed at $p<0.05$.

rFVIIa therapy is efficient for immediate response for those suffering from massive haemorrhage. Patients received rFVIIa were less likely to require further packed cells and blood product transfusion. It was accompanied by a significant improvement in measurement of coagulation parameters. rFVIIa was not associated with incidence of postoperative thromboembolism in this study.

CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND OF THE STUDY

Worldwide population including Malaysians are aware of many health conditions that can lead to disability and death. The situations that usually come across their minds are cancer, heart disease, stroke, physical-related injuries such as road traffic accidents, and infectious diseases. Being least known, bleeding also contributes to one of the common causes of death. Severe cases of uncontrolled bleeding may lead to shock, circulatory failure or serious complication from treatment such as circulatory overload or transfusion related acute lung injury (Sahu et al., 2014) .

Bleeding is a condition that take place when blood leaks from blood vessel and are being drawn out from the circulatory system. A serious condition happens when bleeding occur internally since it is usually being ignored or unrecognised. It can also occur externally through a natural opening such as from the mouth, nose, ear, urethra, vagina, anus, or a break in the skin. Typically, a healthy person can tolerate a loss of 10 to 15% of total blood volume (García, 2009). One who experiences a large amount of blood loss is considered as massive bleeding.

Massive bleeding is a condition where patients need to be transfused with more than 10 units of packed red cells (Meissner and Schlenke, 2012). It is a challenging situation for haematological and blood transfusion services as pressure may arise in order to stop the bleeding, supply blood products and provide laboratory services. Approximately one-third of all bleeding trauma patients are accompanied with a

coagulopathy upon admission to hospital (Frith et al., 2010). These situations increased the incidence of multiple organ failure and death significantly compared to patients with similar injury patterns in the absence of a coagulopathy. According to Spahn et al. (2013), massively bleeding trauma patient needs early identification of bleeding sources, prompt measures to minimise blood loss, restore tissue perfusion and haemodynamic stability.

Apart from mechanical compression at the source of bleeding, transfusion of blood products is one of the standard therapies for any massive bleeding cases. To date, there are a few pharmacological inventions available to facilitate haemostasis such as prothrombin complex concentrate (PCC), tranexamic acid, and recombinant activated factor VII (rFVIIa). However, each choices of treatment has their significant benefits and limitations (Elshamaa and Elokda, 2015). Due to uncertainty about the benefits and risks of these interventions, this study was initiated to survey the outcome of one of these options i.e. rFVIIa in the treatment of massive bleeding.

rFVIIa is a human plasma-derived activated factor VII which was produced by utilising recombinant technology. It is a novel prohaemostatic agent, normally administered intravenously under physicians supervision. It is commercially and widely used in the management of haemophilia patients with inhibitors. To date, the use in non-haemophiliac patients has remained off-label. Some of these off-label indications of rFVIIa are cardiac surgery, trauma, liver transplantation, obstetric haemorrhage and liver cirrhosis has been well-reported (Lin et al., 2012). As a procoagulant agent, the main concern is the risk of thromboembolism. Thus, thus study is also aiming to observe the thromboembolic occurrence among patients who had been treated with rFVIIa.

1.2 RESEARCH OBJECTIVES

1.2.1 General Objective:

- To assess the treatment indications, clinical outcome, transfusion need and coagulation profiles of non-haemophiliac patients receiving rFVIIa during massive bleeding in Hospital Universiti Sains Malaysia (HUSM).

1.2.2 Specific Objectives:

- To study the changes in blood product requirement in 24 hours before and after administration of rFVIIa in massive bleeding patient in HUSM.
- To study the changes of coagulation profile before and after administration of rFVIIa during massive bleeding in HUSM.
- To describe the outcome of mortality and thromboembolic complication of patients received rFVIIa in HUSM.

1.3 STUDY HYPOTHESIS

- Blood product requirements are significantly reduced after administration of rFVIIa in massive bleeding patient in HUSM.
- There is significant improvement of coagulation profiles in massive bleeding patients in HUSM after administration of rFVIIa.
- Massive bleeding patients in HUSM who received rFVIIa have better outcome in term of survival and thromboembolic complication.

1.4 JUSTIFICATION OF THE STUDY

Massive haemorrhage is a possible preventable cause of mortality and morbidity. Thus, it is a serious decision to be make in the massive bleeding management. Correct decision plays an important role in the massive bleeding management. Decision must be quick, proper and accurate because any adverse events may be irreversible.

The optimal resuscitative plan on type of fluid, volume, rate, route of administration, prohaemostatic medications and outcome of resuscitation during massive bleeding remains a debate (Alam and Velmahos, 2011). Sometimes, they may introduce adverse events that are unpredictable or unrecognised post-operatively.

As a consequence, there have been numerous initiatives that aim to improve outcome and prevent complication among the massive bleeding patients. Year by year, haemostatic intervention are evolving and novel drug such as rFVIIa has been introduced.

However, there have been discrepancies in the outcome of rFVIIa among massive bleeding patients. The usage to treat bleeding for non-haemophiliac patients is still debatable. Therefore, this study may contribute to the literature on the outcome of usage of rFVIIa in non-haemophiliac massive bleeding management in Hospital Universiti Sains Malaysia.

To date, most of the data were contributed by the developed countries. The authors aimed to share the research outcome on off-label rFVIIa usage in a developing country ie. Malaysia.

CHAPTER TWO

LITERATURE REVIEW

2.1 MASSIVE BLEEDING

Massive blood loss is defined as loss of one blood volume within 24-hour period. Alternatively, it can also be defined as loss of 50% blood volume within 3 hours or a rate of blood loss of at least 150 ml/min (Stainsby et al., 2000). Major surgery related to cardiovascular, liver, major traumas and oral anti-coagulant therapy are the most common causes of excessive blood loss (Franchini et al., 2008). Treatment strategies of postoperative bleeding comprise of supportive care with volume resuscitation, blood products administration, intervention of pharmacology and surgical re-exploration (Soliman et al., 2012). Failure to recognise bleeding and commencement of an appropriate prompt management is one of the leading cause of preventable death following trauma. A multidisciplinary approach to the prompt recognition of shock due to haemorrhage following trauma is important preferentially before the decompensation.

Massive bleeding events have enormous effect and may potentially deteriorate patients' prognosis and outcomes. Few advancements have been made for understanding of the pathophysiology of trauma, the availability of diagnostic adjuncts and the introduction of new resuscitation protocols (Curry et al., 2011). Even then, the outcome from massive haemorrhage remain poor most of the time in regards to massive transfusion and significant coagulopathy.

Massive bleeding requiring massive transfusion can occur in any clinical conditions and almost always associated with significant mortality and morbidity. Volume

replacement such as fluid and blood product transfusion are the first few essential constituents of the management of patients with massive bleeding. The degree of efficacy of these therapeutic interventions should outweigh potential risks.

2.1.1 Effect of Massive Bleeding

Trauma patients with severe bleeding involve an entangled and crucial management for the medical practitioners. Even though much effort has been put in to understand the pathophysiology of bleeding, the mortality rate remains high. The priority of acute phase of haemorrhage is to arrest the bleeding as quickly as possible by involving fluid resuscitation, blood component transfusion and vasopressors usage (Bougle et al., 2013). According to Meissner and Schlenke (2012), trauma contributes to one of the most common causes of death for individuals between the age of 18 and 45 when it is supposed to be the most productive years in life. In agreement with Curry et al. (2011) and Panteli et al. (2015), this situation represent 40% of mortality and the most common cause of preventable death.

2.1.2 Management of Massive Bleeding

During a severe haemorrhage episode, the top priority is to stop bleeding instantly. If this bleeding is not controlled, there will be continuous deterioration of patient, causing circulatory failure, tissue hypoxia and organ dysfunction. It may be helped by surgery, volume replacement, blood products transfusions and pharmacological interventions(Martinez-Calle et al., 2016).

2.1.2.1 Surgery

Constant haemorrhage in trauma patients may gain benefit from emergency surgery if the elapsed time between injuries to hospital admission is minimized (Cothren et al., 2007). During surgery, the bleeder will be first identified. Clamps and pressure will be applied to slow down the bleeding. If an organ is severely injured causing uncontrolled bleeding, it can sometimes be removed to stop the bleeding, for example by splenectomy, nephrectomy, partial hepatectomy, hysterectomy or even limbs amputation. Subsequently, electrical cautery can be used to burn and stop the bleeding from small vessels. Bleeding vessels can also be identified and tied with sutures (Thomas et al., 2016).

However, some cases of uncontrolled bleeding is unable to be treated by surgery. This include pulmonary haemorrhage and disseminated intravascular coagulopathy due to amniotic fluid embolism or severe sepsis (Shetty et al., 2015).

2.1.2.2 Fluid Replacement

Bouglé et al. (2013) concluded that first therapeutic intervention for haemorrhagic shock is fluid resuscitation. The choice of fluid depends on the patient's conditions and physician's consensus. However, there is no written literature on the supremacy of one type of fluid over another in trauma patients. For example, in a case of brain tumor resection, excessive blood loss may happen. Extensive transfusion of crystalloid or colloid has a tendency of causing volume overload and brain swelling (Vesel et al., 2015b).

Too much of crystalloid or colloid infusion may also lead to dilution of coagulation factors causing consumptive coagulopathy, hypothermia and metabolic acidosis (Nguyen et al., 2009). This may lead to the worsening of the diffuse bleeding.

Crystalloids are solutions of minerals and other water-soluble molecules. Both water and electrolytes will cross semi-permeable membrane into the interstitial space and achieve equilibrium in 2 to 3 hours. 3mL of isotonic crystalloid solution are needed to replace 1mL of patient blood optimally. This is because approximately 2/3 of solution will leave the vascular space in 2 to 3 hours. On the other hand, colloid are large molecular weight solutions with weight of more than 30 kDaltons. They are macromolecular substances made of gelatinous and do not readily cross the semi-permeable membranes. The advantages that colloids have over crystalloids is that the colloids can induce a more rapid and persistent volume expansion because of a greater increase in oncotic pressure (Lundsgaard-Hansen and Pappova, 1981).

Large volumes of crystalloids may relate to tissue oedema, abdominal compartment syndrome (Madigan et al., 2008), and hyperchloremic metabolic acidosis (Handy and Soni, 2008). On the other hand, too much of colloids may easily cause circulatory overload which is hard to be reserved using diuretic medications. Current findings on comparison between colloids and crystalloids in Cochrane review (Perel et al., 2013) reported that there is no solid proof that colloids administration reduced mortality risk as compared to crystalloids in patients with trauma, burns, or after surgery. One possible postulation is, both of these fluid are not the ideal replacement during haemorrhage.

2.1.2.3 Blood and Blood Products Transfusions

Massive bleeding management remains challenging as the patient has high potential to get into haemostatic failure leading to fatal outcome. Alternative strategies to treat bleeding are vital as several approaches proposed to decrease blood loss have

unpredictable degrees of success and complication rate. Basic regime to achieve haemostasis in severe bleeding cases comprise of blood and blood components administration. The conservative treatments include administration of packed red blood cells (pRBC), fresh frozen plasma (FFP), cryoprecipitate or fibrinogen, and platelet concentrates. However, large volume of blood products transfusion are linked to the transfusion adverse reactions such as infections, hypothermia, disseminated intravascular coagulopathy, excessive fibrinolysis, dilutional coagulopathy, and metabolic acidosis, which may worsen bleeding and morbidity (Vasudev et al., 2016). This is supported by Engoren et al. (2002) that transfusion is associated with a 70% increase in disability and death in postoperative cardiac surgery after correction for confounding factors and comorbidities.

Occurrence of transfusion transmitted diseases (TTI) is proven low but the risk is still there especially when there is involvement of numerous number or blood bag transfusion. The most unwanted adverse event of blood transfusion is transfusion-related acute lung injury (TRALI) even though its prevalence is much lower (~1:10,000) than other adverse effects such as febrile or allergy reactions (~1%) (Otsubo and Yamaguchi, 2008).

Early administration of FFP and pRBCs in massive bleeding had been proven to prevent severe dilutional coagulopathy and contributed to survival improvement of patients. However, large amount of further blood components administration may put the patient at risk of Transfusion Associated Cardiac Overload (TACO) (Naveen and Ajju, 2014). 1 to 1.5 units of FFP must be given per unit of pRBC to correct the dilutional component of coagulation alone. Sequel of anaemia, thrombocytopenia, and an increase

of the volume overload may accompanied with imbalance of FFP: pRBC ratio (Spahn et al., 2013).

A study by Hunt (2014) stated that there is still lack of strong quality on evidence of usage of blood components to treat massive bleeding. Administration of blood components routinely in critical care practice from decades ago are still in shortfall since there's lack of study being carried out.

2.1.2.4 Pharmacological Interventions

Advancement of understanding in pathophysiology of haemorrhage has led to improvement of pharmacological interventions for resuscitation strategies. There are a few commonly used drugs in Malaysia which was utilised to treat bleeding with variable safety and efficacy profile. Some interventions are frequently used to uncontrolled bleeding patients, such as tranexamic acid and Vitamin K.

Tranexamic acid is an antifibrinolytic agent. It is a synthetic lysine derivative which acts by inhibiting conversion of plasminogen to plasmin. Thus, it helps to reduce the degree of fibrinolysis. It has been successfully incorporated into most trauma management plans due to its low risk of complication. However, the onset is slow thus it should be given prior to surgery (Kietpeerakool et al., 2016).

Vitamin K or known as phytonadione is also a drug with a haemostasis property. It is commonly used in the prevention of haemorrhagic disease of newborn. It is administered orally or intravenously to reverse the anticoagulant effects of warfarin through promoting the production of clotting factors II, VII, IX and X. Usage of Vitamin K in patients ongoing warfarin treatment with an INR of 4.5 to 9.9 without bleeding is not recommended (Crowther et al., 2009) . It is given by intravenous slowly to avoid severe

hypotension and rare anaphylactic reactions (Riegert-Johnson and Volcheck, 2002). It has a good potential in maintaining coagulation homeostasis in the body. However, due to the slow onset, it is still being used as a supplementary intervention during acute severe bleeding (Marti-Carvajal et al., 2008).

PCC is a concentrated product of plasma with a variety of clotting factors. It was initiated to replace Factor IX in haemophilia B patients. Nowadays, this product is well-known for Vitamin K reversal therapy. It is suggested to use PCC over FFP for patients with massive bleeding while receiving ongoing Vitamin K therapy (Sorensen et al., 2011). However, the product labelling does include a caution of possibilities of developing arterial and venous thrombotic complications associated with its use (Product Information: KCENTRA Intravenous Powder, 2013).

Calcium alginate possesses haemostatic capacity that can be utilised as local haemostatic agent and wound dressing (Qin, 2004). When calcium alginate make a contacts with tissue, calcium and sodium ions are exchanged and form a gel. However, the complications at the injury site is unknown (Taskin et al., 2013).

Aprotinin is one of the powerful antifibrinolytic agents that works as serine protease inhibitor. Its most important effect is to inhibit plasmin, trypsin and kallikreins. It is usually given as prophylactic in patients having vascular surgery to prevent bleeding. However, it is very costly and associated to several side effects such as hypersensitivity reactions related to repeated exposure (Porte and Leebeek, 2002). It is also prone to cause thrombosis to the patients. It was withdrawn from the market since 2008 (Chee et al., 2016) and not available in Malaysia.

Protamine sulphate is used to reverse the effects of heparin but may accompanied with few adverse effects including hypotension, hypersensitivity and paradoxical

anticoagulation with excessive doses (Park, 2004). The potential risk factors for adverse events to protamine included previous exposure, rate of administration, vasectomy and fish allergy (Porsche and Brenner, 1999).

Desmopressin is a synthetic analogue of the antidiuretic hormone vasopressin (Leissinger et al., 2014). Its raises plasma levels of factor VIII (FVIII) and von Willebrand factor (VWF), tissue plasminogen activator (t-PA), and also has vasodilator effect. The release of multimer VWF enhances binding to the subendothelial matrix and platelets. Consequently, increases haemostatic efficacy (Kaufmann and Vischer, 2003). Nevertheless, administration of desmopressin commonly cause headache, facial flushing, mild hypotension and tachycardia. However, many patients treated repeatedly over a short time period become less responsive to these therapy (Franchini, 2007) .

2.2 COAGULATION CASCADE

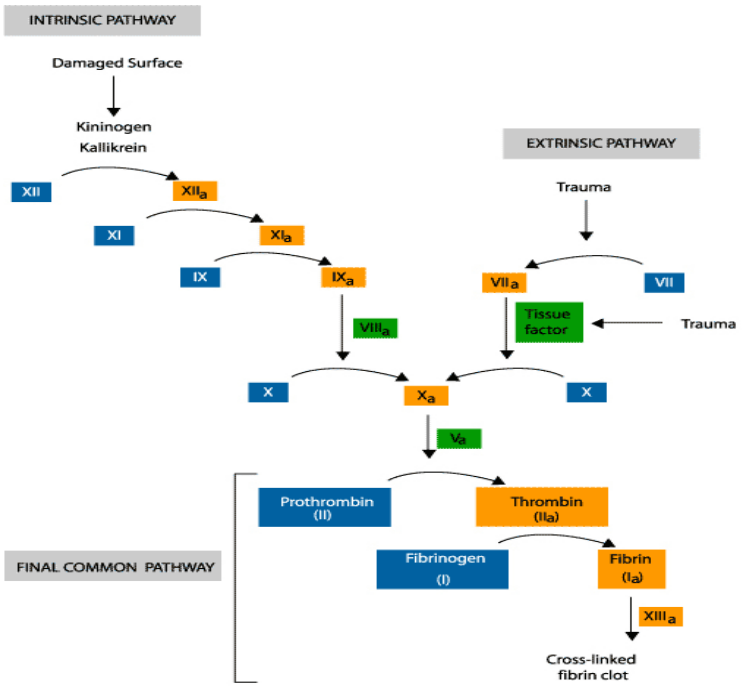

Figure 2.1: Coagulation cascade
(Hoffbrand and Moss, 2011)

Three main parts of coagulation cascade in the body are extrinsic pathway, intrinsic pathway and common coagulation pathway. In case of injury to the blood vessel, tissue factor is brought into contact with naturally occurring activated factor VIIa (FVIIa), forming a complex of TF-FVIIa which then activates FIX and FX. FXa and its co-factor FVa form the prothrombinase complex, which convert prothrombin to thrombin. Conversion of prothrombin to thrombin is the common coagulation pathway. Later, thrombin convert fibrinogen to fibrin to form a localised stable clot (David et al., 2009). The intrinsic pathway initiated with the formation of complex on collagen by high-molecular-weight kininogen (HMWK), prekallikrein, and FXII (Hageman factor). Prekallikrein is converted to kallikrein and FXII becomes FXIIa. Then, FXIIa converts FXI into FXIa. Factor XIa converts FX to FXa and continues with common pathway for clot formation at site of injury (Pallister and Watson, 2010).

2.3 CONFOUNDING FACTORS FOR BLEEDING AND COAGULOPATHY

Different clinical presentation of the patient such as hypothermia and acidosis can contribute to the outcome of haemostasis treatment. Hypothermia and acidosis were reported to influence coagulopathy in different clinical settings .There is significant numbers of mortality in patients with massive haemorrhage with hypothermia, acidosis and coagulopathy. They are known as “lethal triad” (Cosgriff et al., 1997). The outcomes of massive haemorrhage are still poor even though many advancement has been uncovered in regards to the pathophysiology and management of trauma. This may be related to presence of confounding factors in patients. However, the variables are closely linked and cannot be assessed individually in a clinical setting.

2.3.1 Lethal triad

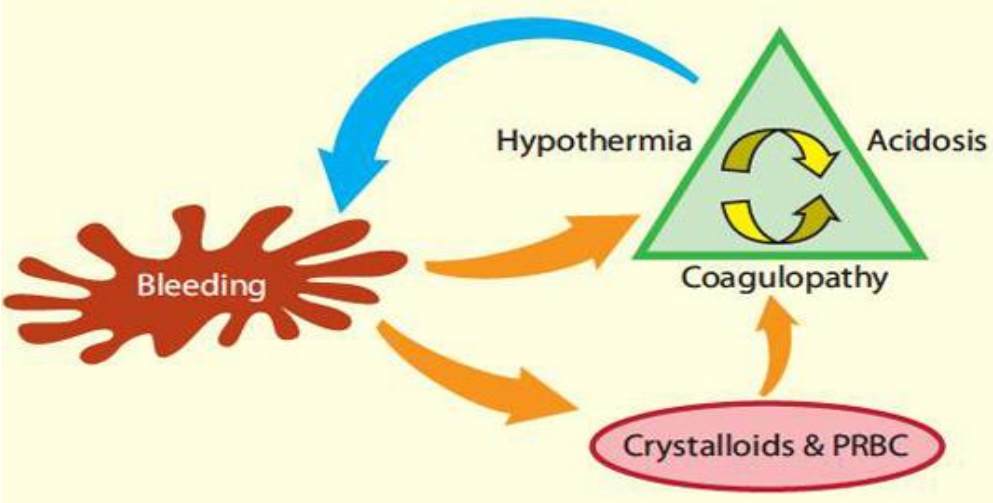

Figure 2.2: Lethal triad, (Gerecht, 2014)

The known "lethal triad" and their well-established contributing role in morbidity and mortality of trauma consists of coagulopathy, acidosis and hypothermia (Smith et al, 2013; Faraoni and Van Der Linden, 2014). Understanding of the pathophysiology of the lethal triad with rapid recognition should serve as the basis for the management of the bleeding trauma patient.

Coagulopathy is any disorder related to the blood coagulation. Coagulopathy related to shock and blood loss has a multifactorial aetiology including consumption of clotting factors, platelets and haemodilution. Additionally, hyperfibrinolysis, hypothermia, acidosis and metabolic changes may also affect the coagulation system. Adrien et al. (2013) reported that the priority goals for early resuscitation of haemorrhage is for the improvement and prevention of coagulopathy. Development of traumatic coagulopathy involves a few mechanism such as consumption of coagulation factors and platelets due to bleeding, haemodilution secondary to overzealous fluid resuscitation and overactivation of coagulation owing to haemorrhage injury. Acute coagulopathy is another contributing factor for mortality and morbidity following trauma (Spahn et al.,

2013). Thrombin generation, fibrin formation and serum fibrinogen concentration are reduced in acute coagulopathy. This may lead to increased transfusion requirement and mortality in bleeding patients (Faraoni and Van Der Linden, 2014).

Acidosis occurred as a result of tissue hypoperfusion and presence of excess chloride. The condition may impair activities of plasma protease, coagulation factor complexes and cell surface interactions. Moreover, acidosis may stunt the thrombin and clotting factors production. Drop of pH from 7.4 to 7.0 decreases the action of factor VIIa by 90%, factor VIIa/ tissue factor complex formation by 55% and factor Xa/Va complex formation by 70%. Acidosis should be avoided as coagulation cascade need an optimal range of pH to function well. For instance, there is 10% a reduction of enzymatic activity of factor VIIa at pH 7.0 instead of 7.4 (Chee et al., 2016). It is also supported by Meng et al. (2003) that reduction in the pH to 7.0 nearly eliminates efficacy of rFVIIa activity.

Hypothermia may cause impairment coagulation in whole blood. The coagulation function is compromised by hypothermia and acidosis synergistically (Dirkmann et al., 2008). Hypothermia can lead to obstructions of protease activity and platelet function. Therefore, upper body warm blankets are used by patients in the emergency room or intensive care unit (ICU) to prevent cooling down (Smith et al., 2013). Additionally, to avoid hypothermia, the transfused intravenous fluids and blood components should be warmed to 37 °C. It is considered as mild hypothermia when the core body temperature is 32°C to 35°C, moderate at 28°C to 32°C, and severe when the core body temperature is below 28°C. Severe trauma related to hypothermia has been associated with 100% mortality. A decrease in temperature can reduce the activity of FVIIa and platelets linearly. Moreover, unconscious or sedated patients are always associated with

hypothermia which leads to reduce in clotting enzyme activity and decrease in platelet function. This process worsens the coagulopathy (Smith et al., 2013).

This triad worsen the haemorrhage and may eventually leads to death. Research suggested correction of hypothermia, acidosis and coagulopathy in trauma is necessary as much as considerations of surgical management. In order to manage critically ill trauma patients, a firm understanding on the lethal triad is essential (Beekley, 2008).

2.3.2 Underlying conditions

Different group of patients may portray different categories of risk factor for bleeding. This include pregnancy, patient taking blood thinning agent, patients with liver failure or patients with hereditary haematological abnormality.

Pregnancy is hypercoagulable state. This condition might be a confounding factor in the treatment with rFVIIa. In normal adults, mean level of plasma FVIIa is 3.6 ng/mL at the range between 0.5 to 8.4 ng/mL with variation between individuals. The levels may be elevated in pregnancy cases and reduced with oral anticoagulant therapy (Fong, 1997). A reproductive studies of rats and rabbits treated with rFVIIa at 6 mg/kg and 5 mg/kg doses revealed that the dose is not associated with mortality and abortion rate. Besides, no sign of teratogenicity was observed after treatment. Nevertheless, treatment of pregnant women with rFVIIa should only be proposed with risk outweigh risk to the foetus. There is also no adverse reaction reported during labour, vaginal delivery and tubal ligation when the doses of 36 μg/kg and 90 μg/kg were given. Excellent response between 80% to 95% in obstetrical and gynaecological situations were observed with rFVIIa administration (Madhusudan et al., 2017).

2.3.3 Previous medication

Patients on treatment with antiplatelet, warfarin or heparin may have an impact on the haemostasis when given rFVIIa. One of the example of antiplatelet is aspirin which believed to cause a significant increase in blood product in cardiac surgery. Aspirin is the drug that is responsible to the interference of platelet aggregation and adhesion. Thus, it modify the haemostasis of postoperative which lead to excessive blood transfusion (Victor et al., 2011). Warfarin which is the Vitamin K antagonist work by inhibiting the vitamin K epoxide reductase. Hence the reusing of inactive vitamin K epoxide back to the active reduced form. This result in exhaustion of active form Vitamin K that is needed for arrest bleeding (Ufer, 2005).

2.3.4 Liver Failure

Hepatic parenchymal cells in liver plays a major role in haemostasis as the place to synthesis the coagulation factors, anticoagulant proteins and components of the fibrinolytic system. The reticuloendothelial system of the liver aids in coagulation and fibrinolysis by eliminating coagulation factors from the circulation. As liver is a highly vascularised organ with vital venous systems draining through the parenchyma, any liver diseases can affect blood flow and cause bleeding problems. Abnormal liver function can resulted from various aetiology such as impairment of coagulation factor synthesis, synthesis of dysfunctional coagulation factors, overconsumption of coagulation factors, deformed clearance of activated coagulation factors and quantitative and qualitative platelet disorders (Faybik et al., 2006).

2.3.5 Hereditary bleeding disorder

The most common hereditary disorder of haemostasis is von Willebrand disease (VWD) while the most common hereditary coagulation disorders are haemophilia. VWD is a heterogeneous haemorrhagic disorder caused by a deficiency or dysfunction of the protein called von Willebrand factor (vWF). Deformed vWF interaction between platelets and the vessel wall meddling with primary hemostasis in the body. In term of coagulation profiles, Haemophilia A and B always has a prolonged aPTT with normal platelets and PT. It is diagnosed with spontaneous joints bleeding, excessive bruising, menorrhagia, unexplained epistaxis and prolonged bleeding after minor cuts, minor surgery or dental procedures (Crookston et al, 2016).

2.4 RECOMBINANT ACTIVATED FACTOR VII

Recombinant activated factor VII (rFVIIa; Novo Nordisk, Bagsvaerd, Denmark) is a novel haemostatic agent act by activating the extrinsic pathway of the coagulation cascade. It was originally developed for the management of bleeding in haemophilia A and B patients with inhibitors to factor VIII or IX by bypassing the intrinsic pathway of coagulation cascade.

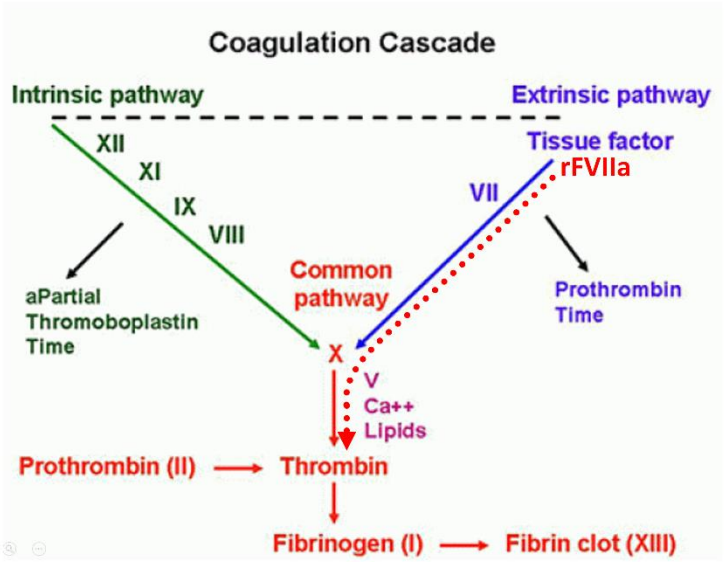

Fig 2.3: Recombinant activated factor VII (rFVIIa) as a bypassing haemostatic agent

It is a vitamin K-dependent glycoprotein consisting of 406 amino acid residues (MW 50 kDa). rFVIIa is secreted into the culture media which contain newborn calf serum in its single-chain form and then proteolytically converted by autocatalysis to the active two-chain form during a chromatographic purification process. The process has been demonstrated to remove exogenous viruses. It is produced by using recombinant DNA technology which is from factor VII cDNA transfected into kidney of baby hamsters and purified by series of chromatography steps, involving immunoaffinity chromatography using murine monoclonal antibodies. It is structurally similar to human plasma-derived Factor VIIa and none of material are extracted from human is used started from production, processes and final product thus it is unlikely to have risk on infectious agents compared to blood products. rFVIIa is presented in glass vials containing 1.2 mg, 2.4 mg and 4.8 mg. It is supplied as a sterile, white lyophilized powder of rFVIIa in single-use vials.

Each vial of lyophilized drug contains the following:

Contents	1.2 mg (60 KIU) Vial	2.4 mg (120 KIU) Vial	4.8 mg (240 KIU) Vial
rFVIIa	1200 μg	2400 μg	4800 μg
sodium chloride*	5.84 mg	11.68 mg	23.36 mg
calcium chloride dihydrate*	2.94 mg	5.88 mg	11.76 mg
glycylglycine	2.64 mg	5.28 mg	10.56 mg
polysorbate 80	0.14 mg	0.28 mg	0.56 mg
mannitol	60.0 mg	120.0 mg	240.0 mg

* per mg of rFVIIa: 0.44 mEq sodium, 0.06 mEq calcium

The action of rFVIIa is limitedly used at the site of injury and exposed tissue factor, thus the risk of systemic activation of coagulation system is almost unlikely to take place (Vesel et al., 2015a). rFVIIa induce haemostasis at optimum doses at injury site by creating complexes with exposed tissue factor and by binding to activated platelet surface. It generates thrombin through direct activation of factor X (Hoffman et al., 1998). It is administered intravenously according to body weight. rFVIIa should not be simultaneously administered with infusion solutions.

2.4.1 Pharmacodynamics

rFVIIa involves in extrinsic clotting pathway where it forms a complex with tissue factor which in the presence of calcium and phospholipids activates coagulation factor X which then initiates the conversion of prothrombin into thrombin at the site of injury (Figure 2.1). Formation of thrombin and clot stabilises platelet plug and form a tight fibrin structure which is resistant to lysis. This process may also occur on the surface of activated platelets.