Introduction
The understanding of blood coagulation has a long history. Even in early days when scientific documents were not there, primitive people realized that loss of blood, if sufficiently great, was associated with death. Hence bleeding to death was one way of punishing people. The emergency resulting due to haemorrhage makes the people to be aware of the importance of blood coagulation.

Coagulation is one of the several defence systems in the blood that maintains the integrity of the circulatory system. In the event of any physical damage, the coagulation system provides an integrated and regulated response, designed to preserve the critical properties of blood volume, pressure, fluidity and integrity.

THROMBOSIS

"Thrombosis means the coagulation occurring in the wrong place or at the wrong time."

The term thrombosis refers to the formation of clot, from the constituents of the blood within the vascular system of a living animal. Thrombosis thus involves the interplay of vascular, cellular and humoral factors within a flowing stream of blood. It is a dynamic process to be distinguished from the static phenomenon of blood coagulation.
Thrombosis is always prevented within the vascular system by many biological mechanisms. This includes,

1. The smoothness of the vascular endothelium
2. A layer of glycocalyx, a mucopolysaccharide on the endothelium which repels the clotting factors and platelets
3. Thrombomodulin, a protein bound with endothelial membrane, when bound to thrombin it slows the coagulation process and also activates the plasma protein C, which in turn, inactivates the activated Factors V and VIII.

Apart from this, the natural anticoagulant heparin when bound to antithrombin III increases the effectiveness of antithrombin III activity in removing thrombin by 100 to 1000 folds and thus it acts as an anticoagulant (Guyton and Hall, 2001).

In the recent days, thrombosis together with complicating embolic phenomena in the veins and arteries is most important cause of sickness and death in the developed and developing countries. The combination of myocardial infarction and thrombotic stroke consistently represents the major cause of death in the United States, numbering over 800,000 people annually (Parker et al., 1996). The mortality rate with acute myocardial infarction (AMI) is approximately 30% with more than half of these occurring even before the affected individual reaches the hospital. The incidence of deep vein thrombosis and pulmonary embolism in Asians, Africans and Chinese is also reported (Braunwald et al., 2001).
PATHOPHYSIOLOGY OF THROMBOSIS

Excessive activation of coagulation or inhibition of anticoagulant mechanisms may result in hypercoagulability and thrombosis. The abnormalities of the vessel wall, alterations of blood flow and changes in the composition of the blood are the major factors in the pathophysiology of thrombosis, and were well recognized in the nineteenth century. The haematologic aspects of thrombosis are considered here for these three factors, are usually referred to as the triad of Virchow (Figure I).

Vascular injury

Arterial thrombosis often is the result of a process that damages the vessel wall, such as atherosclerosis or homocysteinemia. When the endothelial wall damages, the smoothness and its glycol-calyx-thrombomodulin layer are lost, thus activate the factor XII and platelets. It commonly begins with platelet adhesion on an abnormal vascular surface and formation of a nidus of platelets and fibrin. This process may be potentiated by adenosine diphosphate (ADP) or other mediators that are released from activated platelets (Ross, 1993). The release reaction and platelet synthesis of thromboxane A2 (TXA2) and other agonists induce the aggregation of more platelets which causes enlargement of the thrombus, a phenomenon often called as ‘platelet recruitment’.

In venous thrombosis, the vessel wall is usually histologically normal, factors extrinsic to the vessel appear to have the major pathophysiologic role. An exception to this generalization is direct venous trauma or venous vascular
Figure - I  Pathophysiology of arterial and venous thrombosis

Richard et al, 1999
disease. A generalized reduction in venous tone may be an important pathophysiologic factor in venous thrombosis.

**Abnormalities of Blood Flow**

Arterial thrombosis may be contributed by turbulent blood flow and hyperviscosity of the blood. Clots that formed in arteries under conditions of high blood flow are predominantly composed of platelets and have little fibrin. These *white thrombi* may readily dislodge from the arterial wall and embolize to distant sites, causing temporary or permanent ischemia. These clots are particularly common cause of embolism in the cerebral and renal circulation where they may lead to transient neurological dysfunction (transient ischemic attacks) including temporary mono ocular blindness (amaurosis fugax) or stroke. In addition, most episodes of myocardial infarction are due to thrombi that form after the rupture of atherosclerotic plaques within the diseased coronary arteries.

Venous thrombosis develops under conditions of slow blood flow, and is augmented by further retardation and stagnation of the flow. Haemostatic plug or thrombin that forms in vein where the blood flow is slow which is rich in fibrin, trapped RBC and containing relative few platelets. They are often called *red thrombi* because of their appearance in surgical and pathological specimens. The friable ends of these red thrombi which most often form in leg veins can break off and embolize to pulmonary circulation (Richard *et al.*, 1999).
Coagulation Abnormalities

Elevated levels of various coagulation factors, particularly fibrinogen and factors V, VII, VIII, and X, have been documented as the cause of thrombosis and pre-thrombotic disorders. Fibrinogen and factors V and VIII are acute-phase reactants, and their plasma levels may rise in patients with virtually any disorder associated with tissue damage or inflammation, including most thrombotic processes. On the other hand, recent epidemiological studies have identified elevated plasma levels of fibrinogen and factor VII activity as independent risk factors for cardiovascular disease. An association between elevated plasma von Willebrand factor (vWF) levels and recurrent myocardial infarction has been demonstrated, and vWF antigen is an independent predictor of coronary artery disease (Meade et al., 1986).

Abnormal fibrinolysis has also been linked to vascular diseases. Low fibrinolytic activity was a significant determinant of coronary artery disease and elevated plasminogen activator inhibitor-1 (PAI-1) activity was associated with major ischemic events (Meade et al., 1993).

The thrombotic disorders can be described in two categories (Richard et al., 1999)

I. Inherited thrombotic disorders

II. Acquired disorders predisposing to thrombosis
I. INHERITED THROMBOTIC DISORDERS

Many people affected with thrombosis are found to have inherited defects that are combination of deficiency or qualitative abnormalities of activated coagulation factors. The different types of inherited thrombotic disorders are,

1. Antithrombin III deficiency

Antithrombin III (ATIII) deficiency was described by Egeberg in 1965. Like most inherited thrombotic disorders, ATIII deficiency is inherited as an autosomal dominant disorder. A blood bank survey reported that 1 in 600 people have ATIII deficiency. ATIII deficiency is manifested primarily by recurrent venous thromboembolism. Almost every vein site has been reported to be involved with thrombosis in ATIII-deficient patients, including unusual sites such as mesenteric vessels (Marciniak et al., 1974).

2. Protein C deficiency

Inherited deficiency of protein C and its association with thrombosis was first described by Griffin and coworkers in 1981 as an autosomal dominant disorder. The predominant clinical symptom of protein C-deficient patients is recurrent venous thromboembolism, although arterial thrombotic events, including stroke have been reported (Camerlingo et al., 1991).

3. Protein S deficiency

Protein S is a vitamin K dependent protein that mediates the anticoagulant activity of activated protein C. Protein S deficiency in
association with inherited thrombolytic disease was first described by Comp and Esmon (1984). Like ATIII and protein C deficient patients, most patients with protein S deficiency and thrombosis will experience venous thromboembolism.

II. ACQUIRED DISORDERS PREDISPOSING TO THROMBOSIS

1. Vascular disorders

Atherosclerosis
Diabetes
Vasculitis
Prosthetic materials (grafts and valves)

2. Abnormal rheology

Stasis - immobilisation, surgery, congestive heart failure
Hyperviscosity- polycythemia vera, acute leukemia, sickle cell disease

3. Platelet dysfunction

Myeloproliferative disorder
Paroxysmal nocturnal hemoglobinuria

4. Other disorders associated with hypercoagulability

Cancer
Oral contraceptives and estrogen therapy
Pregnancy
Infusion of prothrombin complex concentrates
Nephrotic syndrome
Thrombotic thrombocytopenic purpura
Disseminated intravascular coagulation
Antiphospholipid antibody syndrome
Heparin-induced thrombocytopenia/thrombosis
Autoantibodies to calpastatin
Organ transplant

ANTITHROMBOTIC THERAPY

Thrombotic disorders typically accounts for 30 to 40% of all deaths annually in the United States (Parker et al., 1996). Nonfatal thrombotic disorders, including venous thrombosis and transient ischemic attacks affect hundreds of thousands of additional patients every year. The therapeutic modalities available for prophylaxis and treatment of thrombotic disorders are anti thrombotic or thrombolytic drugs. The pathological mechanism of arterial and venous thrombosis forms the basis for the mode of treatment, which includes antiplatelet or antithrombin activity, or lysis of fibrin clots.

The well-established antithrombotic drugs towards the prophylaxis and treatment of thrombotic disorders are aspirin, heparin and warfarin. Two major indications for heparin are,

1. It is used for prophylaxis and towards the treatment of thromboembolism, arterial thrombosis, deep vein thrombosis and anticoagulation following thrombolytic therapy or angioplasty for myocardial infarction and unstable angina.
2. It is used to maintain catheter patency for dialysis procedure and as an anticoagulant for extracorporeal circulation (cardiopulmonary bypass) (Richard et al., 1999).

Warfarin is the most widely used coumarin derivative oral anticoagulant and many consider it to be drug of choice. The therapeutic action depends upon the ability to prolong the prothrombin time by suppressing the synthesis of prothrombin and factors VII, IX and X by competitive inhibition of vitamin K in the liver. Aspirin which interferes with platelet aggregation is useful in the prevention and treatment of thrombosis (Satoskar and Bhandarkar, 2001).

THROMBOLYTIC THERAPY

Thrombolytic agents are in widespread use for the dissolution of arterial and venous pathologic thrombi. Unlike other antithrombotic agents, these types of thrombolytic agents are effective at lysing fibrin clots. Clinical conditions where thrombolysis plays an important role include the acute coronary syndromes, peripheral arterial occlusion, ischemic stroke, deep venous thrombosis, and pulmonary embolism. Thrombolytic agents have been successfully employed in each of these areas, achieving dissolution of the occlusive thrombus, reconstitution of blood flow, and improvement in the status of the tissue bed supplied or drained by the involved vascular segment. All clinically available thrombolytic agents act through the cleavage of the plasminogen molecule to its active form, plasmin (Kenneth, 2002).
The major thrombolytic agents in clinical use include streptokinase (SK), urokinase (UK), and tissue plasminogen activator (t-PA). Apart from these three, plasminogen activator (PA), reteplase (rt-PA), anistreplase (APSAC), and tenecteplase (TNK-t-PA) have been approved by US Food and Drug Administration (FDA) for use of major thrombotic diseases. The major distinction between PAs is related to their origin and antigenicity, half-life, potential for inducing a lytic state and heamorrhagic potential (Marder, 2001).

CLINICAL EFFICACY OF THROMBOLYTIC THERAPY

The clinical efficacies of antithrombotic and thrombolytic therapy in treating ischemic heart disease, venous thromboembolism, cerebrovascular disease and peripheral vascular disease have been well established (Collins et al., 1997).

ACUTE MYOCARDIAL INFARCTION

Acute myocardial infarction (AMI) is one of the most common diagnoses in hospitalized patients in industrialized countries. In United States approximately 1.1 million AMIs occur each year. The WHO has drawn attention to the fact that Coronary Heart Diseases (CHD) is the disease that affects population not unavoidable attribute of ageing. Myocardial infarction is specific to CHD (Parker, 2000).

AMI generally occurs when coronary blood flow decreases abruptly, after a thrombotic occlusion of a coronary artery previously narrowed by atherosclerosis. AMI occurs when a coronary artery thrombus develops
rapidly at a site of vascular injury. The principle goal of thrombolysis is prompt restoration of coronary arterial patency. Thrombolytic therapy can reduce the relative risk of in-hospital death by up to 50% when administered within the first hour of the onset of symptoms of AMI, and much of this benefit is maintained for at least ten years. Approximately, uses of thrombolytic therapy appears to reduce infarct size, limits left ventricular dysfunction, and reduces the incidence of serious complications such as septal rupture, cardiogenic shock and malignant ventricular arrhythmias. (Braunwald et al., 2001).

DEEP VENOUS THROMBOSIS

The prevalence of venous thrombosis is particularly high in patients with cancer of pancreas, lungs, genitourinary tract, stomach and breast. Approximately, 10 to 20% of patients with idiopathic deep venous thrombosis have or develop clinically overt cancer. The most important consequences of this disorder are pulmonary embolism and syndrome of chronic venous insufficiency.

Early administration of thrombolytic drugs may accelerate clot lysis, preserve venous valves, and decrease the potential for developing postphlebitic syndrome. Thrombolytic therapy in Pulmonary Thromboembolism (PTE) may rapidly reverse right heart failure and thus lead to a lower rate of death. Thrombolysis achieves these stated results by dissolving much of the anatomically obstructing pulmonary arterial thrombus and dissolving much of the source of thrombus in the pelvic or deep leg veins thereby decreasing the likelihood of recurrent PTE (Braunwald, 2001a).
ACUTE CEREBRAL INFARCTION

Stroke is the third most common cause of death in developed countries which is uncommon below the age of 40 years and more common in males. Thromboembolism leads to acute cerebral infarction and stroke. The use of thrombolytic agents in acute cerebral infarction has been studied extensively. The National Institute of Neurological Disorders and Stroke (NINDS), USA showed that a clear benefit of recombinant t-PA (rt-PA) in selected patients with acute stroke. If patients are carefully selected and treated within 6 - 7 hrs, neurological sequelae of stroke are reversible (Kumar and Clark, 1998).

CONTRAINDICATIONS OF THROMBOLYTIC DRUGS

Clear contraindication to the use of thrombolytic agents include a history of cerebrovascular haemorrhage at any time, a nonhaemorrhagic stroke or other cerebrovascular event within the past one year, marked hypertension and active internal bleeding (excluding menstruation).

Relative contraindications to thrombolytic therapy are,

1. Current use of anticoagulant
2. Cardio pulmonary resuscitation
3. Known bleeding diathesis
4. Pregnancy
5. A haemorrhagic ophthalmic conditions
6. Active peptic ulcer disease
Complication of currently available thrombolytic drugs

Bleeding is the major complication of the thrombolytic therapy. These drugs cannot distinguish between the fibrin of pathologic thrombi and fibrin in haemostatic plugs present, for example in the gastric or cerebral vasculature. This nonselective fibrinolytic action, coupled with lack of predictive laboratory test for bleeding makes it mandatory to carefully evaluate patients before initiating thrombolytic therapy. Bleeding occurs often at sites of recent surgery or invasive procedures. Intracranial bleeding occurs in approximately 1% of treated patients, and is independent of thrombolytic agent when the drugs are used in their recommended therapeutic dosages. Large-scale intervention trials have suggested that the rate of intracranial haemorrhage with t-PA or rt-PA is slightly higher than that with streptokinase.

Allergic reactions to streptokinase occur in approximately 2% of patients who receive it. When a minor degree of hypotension occurs in 4-10% patients given with this agent, marked hypotension occurs, although rarely, in association with severe allergic reactions (Braunwald, 2001).

Search for new thrombolytic agents

The quest for new thrombolytic agents with a higher thrombolytic potency, specific thrombolytic activity and better fibrin selectivity continues.
Several lines of research towards improvement of thrombolytic agents are being explored as the need for such a therapy is essential.

One of such research includes Thrombinase, a new thrombolytic enzyme which has been developed from a bacterial source and studied for its efficacy and unwanted reactions in animal models.