This review concentrates on discussing the various therapeutic agents available to prevent or inhibit clot formation. Particular emphasis is placed on therapies associated with modification to coagulation factors, and the inhibitors of thrombin formation and action. The genesis of ischaemic cardiovascular disease is related to inappropriate platelet function and/or thrombin generation in excess amounts or at inappropriate sites. The most commonly used agents currently available to inhibit or slow thrombin production include vitamin K antagonists and heparin, acting through circulating or endothelial-derived intermediaries. More recently, a number of agents, which can act to directly inhibit thrombin, have been licensed for use in humans. It is expected that the range of these compounds will eventually grow to replace the use of unfractionated heparin (UFH) and vitamin K antagonists within the next few years. To better understand the mechanism of action of all of these compounds, a brief description of the mechanism of clot formation and the pivotal role of thrombin in this process is required together with the mechanisms for localizing and controlling this activity.

The coagulation cascade

The aim of the coagulation phase of haemostasis is the generation of fibrin strands that will bind and stabilize the weak platelet haemostatic plug. There are no covalent bonds holding the platelets together during the formation of the primary haemostatic plug. If left in this state the platelet plug, formed by platelet aggregation, would come apart in a few hours, resulting in late bleeding. The process of blood coagulation, with soluble factors in the blood entering into a chain of reactions that lead to the formation of fibrin, is intended to be localized to the area where the original platelet plug was formed.

This localization is achieved by two methods. First, the chain of reactions which led to the conversion of fibrinogen to fibrin are programmed to occur, and are most efficient and explosive, when restricted to a surface, such as platelet phospholipid. Second, there are a series of inhibitors that are intended to constrain the reaction to the site of injury and platelet deposition. These inhibition processes include the following.

1. Circulating factors such as antithrombin III (ATIII) and heparin cofactor II (HCII).
2. Those derived from endothelium such as tissue factor pathway inhibitor (TFPI).
3. The thrombomodulin system, which converts prothrombotic thrombin to an anticoagulant through the activation of circulating protein C.

Localization of the coagulation process

Historically, the blood coagulation system is divided into two initiating pathways: the tissue factor (extrinsic) pathway and the contact factor (intrinsic) pathway which meet at a final, common pathway, whereby factor Xa converts prothrombin to thrombin which then acts on fibrinogen. These pathways were identified and categorized during experiments to examine the effects of sufficiency and deficiency of the various circulating factors on assays of plasma coagulation. At present the immediate clinical investigation of haemostatic disorders still requires this compartmentalized, cascade-type model as the laboratory-based tests of coagulation focus on each of these separate aspects. The prothrombin time (PT) is a plasma and test-
Tube variant of the extrinsic pathway, and the activated clotting time (ACT) or activated partial thromboplastin time (aPTT) of the intrinsic system for blood and plasma, respectively.

This model based on the concept of a waterfall or cascade is an over simplification of the system, as proteins from each pathway can influence one another. It is probably more correct to think of the coagulation system as an interactive network with carefully placed amplifiers and restraints.

Fibrin formation is a process of initiation and amplification. The specific properties of platelets and the coagulation system cooperate to ensure that fibrin formation occurs only at the localized site where it is required to initiate wound repair. This is achieved by a number of physico-chemical means.

The surface of resting platelets contains acidic phospholipids such as phosphatidylserine that have their negatively charged pole directed inward. Spontaneous reversal of this charge is countered by a specific enzyme system in the platelet (a lipase), implying that this charge reversal is of pivotal importance. When the platelet becomes activated, the negatively charged phospholipid remains on the outside surface of the platelet membrane and is not flipped internally.

The coagulation system relies primarily on a group of soluble factors that circulate in the plasma. These factors are synthesized in the liver and expressed into the circulation (Table 1). Most coagulation factors are identified by Roman numerals, the active form denoted by the lower case ‘a’. They circulate in an inactive, zymogen form and become active after proteolytic cleavage. The exception to this is factor VII that can circulate as an active protease. Apart from factor XIII, which is a transglutaminase, all the active factors are serine protease related to the digestive enzyme, trypsin. Other factors in the coagulation process, such as tissue factor, factor V, factor VIII, and high molecular weight kininogen (HK) act as co-factors.

Factors VII, IX, X, and prothrombin depend on the presence of vitamin K for their conversion to a protein which can optimally participate in the generation of thrombin. These vitamin K-dependent coagulation factors possess groups of negatively charged glutamic acids at their N-terminal regions. Vitamin K acts as a co-factor for an enzyme that adds carboxylic acid to the glutamic acid, forming gamma-carboxy glutamic acid, with a resultant higher density of negative charges localized to the N-terminus. Most of the vitamin K-dependent factors have between nine and 12 of these gamma-carboxy glutamic acid groups available for reaction. This charged area of the polypeptide is the part of the molecule that binds or attaches to the organizing surface of the platelet (Fig. 1).

It is obvious that the highly negatively charged surface of the activated platelet, produced by the expression of acid phospholipids, and a protein that is also negatively charged, will not come together. The role of calcium ions, with their positive charge, is to act as a buffer or sandwich between these areas of charge. In addition, calcium can induce a conformational change in the coagulation protein to enhance or enable binding to the surface of the activated platelet. This process has been likened to landing on the deck of an aircraft carrier. The conformational change produced by calcium will cause the vitamin K-dependent protein to drop its ‘tailhook’, which can then catch the ‘wire’ on the activated platelet surface. By this process, the coagulation proteins arrest and stop at the site of the injury.

<table>
<thead>
<tr>
<th>Standard nomenclature</th>
<th>Traditional name</th>
<th>Molecular weight (Da)</th>
<th>Plasma concentration (μg ml⁻¹)</th>
<th>Half-life (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Factor I</td>
<td>Fibrinogen</td>
<td>340 000</td>
<td>2000–4000</td>
<td>90</td>
</tr>
<tr>
<td>Factor II</td>
<td>Prothrombin</td>
<td>72 000</td>
<td>120</td>
<td>65</td>
</tr>
<tr>
<td>Factor III</td>
<td>Tissue factor</td>
<td>45 000</td>
<td>0</td>
<td>–</td>
</tr>
<tr>
<td>Factor IV</td>
<td>Calcium</td>
<td>40</td>
<td>100</td>
<td>–</td>
</tr>
<tr>
<td>Factor V</td>
<td>Proaccelerin</td>
<td>330 000</td>
<td>10</td>
<td>15</td>
</tr>
<tr>
<td>Factor VII</td>
<td>Proconvertin</td>
<td>48 000</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Factor VIII</td>
<td>Antihaemophilic factor</td>
<td>360 000</td>
<td>0.05</td>
<td>10</td>
</tr>
<tr>
<td>Factor IX</td>
<td>Christmas factor</td>
<td>57 500</td>
<td>4</td>
<td>25</td>
</tr>
<tr>
<td>Factor X</td>
<td>Stuart-Prower factor</td>
<td>55 000</td>
<td>12</td>
<td>40</td>
</tr>
<tr>
<td>Factor XI</td>
<td>Plasma thromboplastin antecedent</td>
<td>160 000</td>
<td>6</td>
<td>45</td>
</tr>
<tr>
<td>Factor XII</td>
<td>Hageman factor</td>
<td>85 000</td>
<td>40</td>
<td>50</td>
</tr>
<tr>
<td>Factor XIII</td>
<td>Fibrin stabilizing factor</td>
<td>320 000</td>
<td>20</td>
<td>200</td>
</tr>
</tbody>
</table>
Coagulation organization: amplification for explosive thrombin production

A recurrent theme in the coagulation system is the formation of activation complexes involving a serine protease, a zymogen or substrate, a co-factor, and an organizing surface, usually provided by the platelet membrane. These factors must be presented to each other in a tightly controlled way to ensure the process of coagulation is amplified and progressed with sufficient rapidity. This elegant organization can be appreciated by considering the formation of thrombin from prothrombin, which is the best characterized of such reactions.

It is axiomatic that production of thrombin needs to be explosive at the site of the injury in order to prevent it being washed away to cause havoc elsewhere in the vascular tree. The major problem to overcome in this process is to enable factor Xa to remove the restraint or protective bubble over the active site on prothrombin at a sufficient rate.

Acceleration of the process is achieved by maximizing the encounter of active site and substrate by appropriate orientation of each molecule (Fig. 2A).

Factor V overcomes this problem by engaging itself into the organizing surface and then holding the two factors by a ‘handle’ to align the proteins in the required configuration (Fig. 2A). This surface-bound enzymatically active system is called the ‘prothrombinase complex’. An identical series of reactions occurs to produce factor Xa. In this case, the non-vitamin K-dependent co-factor VIII reaches up from its binding site on a platelet surface and grabs both factor IXa (the serine protease) and factor X (the zymogen), aligning them correctly for maximum interaction and generating factor Xa. The factor IXa, VIII, and X complex is referred to as the ‘tenase’ and factor Xa, V, and II as the ‘prothrombinase’ complex (Fig. 3).

Factor V is synthesized in the liver and has a half-life of about 5–6 h in stored blood and about 15 h in circulating plasma. In addition to the 10 μg ml⁻¹ circulating in plasma, a significant reserve of factor V is contained in platelets. This platelet factor V adds a further 25% to the circulating pool and is released to the cell surface when the platelet is stimulated by a variety of agonists. Factor VIII is a large unstable molecule that circulates in complex with von Willebrand Factor (vWF). Plasma half-life without the vWF is 2.5 h compared with about 10–12 h with this co-factor.

The pivotal importance of these co-factors (V and VIII) becomes obvious when the kinetics of the reactions were considered. If we assume first that the rate constant for the conversion of prothrombin to thrombin by factor Xa in an aqueous solution is unity, then addition of calcium ions to this mixture will make the reaction rate 2.3 times faster. Activation of prothrombin by factor Xa is 22 times more likely to occur with both factor Xa and prothrombin combining on the negatively charged surface of platelet phospholipid than if the enzyme and substrate were just floating in solution. The addition of factor V to this mixture,
by holding factor Xa and prothrombin in place and aligning them correctly, so they have no choice but to have a productive interaction, accelerates the reaction by 278 000-fold.

If similar kinetics are assumed for the activation of factor Xa, then simple mathematics show a colossal 7.7 million-fold acceleration of thrombin generation in a fully active system. Put another way, a fluid phase two-step coagulation process for the generation of thrombin, involving first the activation of factor X to Xa followed by the activation of prothrombin to thrombin would require about 3 months (89 days). This same reaction would take only 1 s in the unrestricted fully intact system. If the process works properly then explosive thrombin formation should occur only where it is directed and required.

**Tissue factor and thrombin generation**

Factor Xa can also be generated by a different, surface-dependent, pathway. The so-called tissue factor or extrinsic pathway is considered to be the principal initiating pathway of coagulation in vivo. In this reaction, factor X is cleaved by the serine protease, factor VII. Factor VII is held in the appropriate conformation by endothelial surface-bound tissue factor (Fig. 4).

Tissue factor (TF) is a glycoprotein consisting of an extracellular, transmembrane, and cytoplasmic domain that is not observed free in the circulation. This factor is normally only expressed at sites physically separated from circulating blood such as the subendothelium of blood vessels, organ capsules, cells of epithelial surfaces, and the nervous system. Animal brain tissues are the usual source of tissue thromboplastin used for stimulation of coagulation in the PT test. The fact that TF requires phospholipid for full activity and that it has a large transmembrane domain help to retain the TF:factor VIIa:factor X activating complex at the cell surface, ensuring that coagulation is localized to the site of injury. The factor Xa formed remains bound in the surface phospholipid and forms the prothrombinase complex with factor V and prothrombin.

Circulating factor VII is an unusual coagulation protein for two reasons: (1) the non-activated (zymogen) form has some proteolytic activity and (2) about 1% of the circulating form exists as the active enzyme, factor VIIa. Either form can bind readily to TF and the complex thus formed will have enough activity to cleave factor X to factor Xa. Factor Xa will then rapidly convert the factor VII:TF complex to factor VIIa:TF and can thus potentiate the reaction. In addition to activating factor X, the factor VIIa:TF complex can also cleave factor IX to form factor IXa, which can then itself activate factor X as described above. This illustrates the lack of division of the contact and TF pathways in vivo.

Peripheral blood cells do not normally express TF. However, circulating monocytes and endothelial cells can be induced to produce TF by a variety of stimuli including endotoxin, tumour necrosis factor, interleukin 1, immune complexes, hypoxia, and hypothermia. This stimulation and expression of TF by phlogistic agents is thought to play a part in the consumptive coagulopathy associated with sepsis.

**Thrombin and its activity**

Thrombin is a serine protease that has become the focus of considerable research interest. This interest is driven by observations of the ubiquitous actions of thrombin and also by advances in our understanding of the molecular mechanisms involved in the structure and activity of this protease. In turn, this has lead to the development of agents that have either a direct effect on the cleavage site of the molecule or can in some other way inhibit the ability of thrombin to catalyse the conversion of fibrinogen to fibrin.

The thrombin molecule can be simplistically viewed as a sphere with a $G$ shaped groove along its equatorial axis (Fig. 5). The horizontal part of the groove extends around the molecule. The left-hand part of the horizontal section is included with the vertical area to make up the active proteolytic site. The extended horizontal groove, distal to the active site, is one of a number of anion binding exosites on the surface of the thrombin molecule. This specific exosite of thrombin is important as it is involved with thrombin inhibition/binding by the heparin: ATIII complex and also the carboxyl tail of hirudin (see below).

Thrombin cleaves fibrinogen by the removal of two small polypeptides (termed fibrinopeptide A and B, respectively) to expose a site in the centre of the molecule. This is able to tether to the bulbous ends of other fibrinogen, or fibrin,
molecules made from the β and γ chains. The longer α chain of the fibrin molecule projects from the bulbous end and acts to wrap the polymer and protect the binding sites. The fibrinogen molecule has a number of fascinating aspects to its chemistry and evolution. In particular, with reference to this review, is that about 40% of thrombin is incorporated into fibrin during its formation from fibrinogen. This thrombin is thus protected from natural inhibitors.\(^{25, 49}\)

Inhibitors of coagulation in humans

The process of thrombin generation must be localized and contained to prevent global thrombosis after minor injury. It is a basic tenet in biochemistry that every activator will have a cognate inhibitor and this is true for the coagulation system. The natural inhibitors fall into two main groups, endothelial or hepatic, based on their synthetic site. An alternate classification could be to separate the inhibitors into those that aim to inhibit thrombin production and those that directly inhibit this enzyme.

**Endothelial-derived inhibitors**

Endothelial-derived factors include TFPI and activated protein C (aPC). This latter protease has been investigated as a means of preventing thrombus formation or extension. The association with aPC and its generation are 2-fold. First, thrombin is responsible for the generation of aPC. Thrombin is held on the endothelial surface by a co-factor/receptor called thrombomodulin. The active site of the thrombin cleaves the protein C moiety to release aPC (Fig. 6). This clever device allows thrombin to be converted from a procoagulant to an anticoagulant protein. aPC is a serine protease that cleaves a peptide from the arms of factor V and factor VIII, thereby preventing appropriate participation in the tenase and prothrombinase complex (Fig. 7). Resistance to this cleavage is observed in patients who have a single point mutation in their factor V (so-called factor V Leiden). Similar to a genetic absence of protein C, this is not a lethal gene. However, patients with the Leiden mutation are at substantially increased risk of venous thrombosis,\(^4\) and myocardial infarction in certain populations.\(^{66}\) The second interest in the relationship between thrombin and protein C lies in the evolution of these proteins. Genomic mapping suggests that these proteins developed and evolved together. We also increasingly recognize a functional relationship between these proteins. One example is that cleavage by thrombin, to release the tethered ligand of the thrombin receptor, occurs at a site with structural and amino acid sequence homology with protein C.

TFPI is a relatively new addition to the ranks of the inhibitors of coagulation.\(^{65}\) Human TFPI is a protease inhibitor that consists of three Kunitz-type serine protease inhibitor like domains (K1, K2, and K3) flanked by a negatively charged N-terminus and a positively charged C-terminal tail. TFPI is synthesized and released from the endothelium and appears to be the main inhibitor of the TF pathway \textit{in vivo}. The majority of TFPI (85%) is bound to the endothelium and can be rapidly released on stimulation. The remaining 15% is associated with plasma lipoproteins accounting for its early title of lipoprotein-associated coagulation inhibitor. Thrombin production is inhibited and slowed as follows. The second domain (K2) of TFPI binds (reversibly) to the active site of factor Xa inhibiting this protease (Fig. 7). The K1 domain then interacts with the TF:factor VIIa catalytic complex forming an inactive complex.\(^{28}\)
Individuals with congenital TFPI deficiency have not been identified. Mice homozygous for deletion of K1 in the TFPI gene die in utero implying that such a deficiency is not compatible with life. Of interest is that the inhibitor site of the K1 domain differs by only 1 amino acid residue from the inhibitor Kunitz domain of aprotinin.

**Inhibitors by hepatic synthesis**

These circulating factors include a number of serine protease inhibitors or serpins. This superfamily of proteins plays a major role in the regulation of coagulation, fibrinolysis, and inflammation. The serpins function as suicidal inhibitors, presenting their reactive centre as a pseudo-substrate for their target. Although hydrolysis is attempted by the protease it cannot be completed and a tight 1:1 complex is formed which is rapidly cleared from the circulation. For example, the half-life of the thrombin–ATIII (TAT) complex is about 5 min. The two plasma inhibitors found most commonly are ATIII, which accounts for about 60% of plasma anticoagulant activity, and HCII which accounts for a further 30% of the total activity.

ATIII is synthesized in the liver and is not vitamin K-dependent. This inhibitor irreversibly neutralizes factors Xlla, Xla, IXa, Xa, and thrombin (Fig. 7). In vivo, glycosaminoglycans such as heparan sulphate on the endothelial cell surface are the initiators of the enhanced ATIII inhibitory function. Heparan is a varying chain length mucopolysaccharide or glycosaminoglycan that is tethered to the surface of the endothelium by a protein skeleton. Contact with this surface will induce a conformational change in ATIII. This combination produces the physiological effect of a vascular surface with profound anticoagulant properties.

ATIII has a biological half-life of 3–5 days and is produced at a relatively constant rate. It is not an acute phase respondent and so production does not change rapidly in response to stress. Deficiency of ATIII can be congenital or acquired. The normal range for ATIII is based on a...
comparison with pooled plasma and is quoted as 85–120%.
The congenital forms can be divided into those associated
with an absence or reduction of ATIII in the plasma and
those associated with an amino acid sequence that bestows
inappropriate inhibitory activity to the molecule. Both these
defects are associated with an increased risk of thrombo-
embolic disease.

Acquired ATIII deficiency is seen in a number of states
including certain chemotherapeutic regimens (L-asparagin-
ase treatment), hepatic failure, nephrotic syndrome, severe
pre-eclampsia, shock, disseminated intravascular coagula-
tion (DIC), and after certain surgeries such as those
involving extracorporeal circulation. ATIII deficiency is
also seen in chronic heparin administration, to produce
‘heparin resistance’.

Pregnancy represents an interesting example and model
of reduced ATIII activity that is relevant to other clinical
arenas. Fibrinogen concentration and platelet count increase
during pregnancy, and in pre-eclampsia there are diminu-
ted levels of ATIII. This fall in ATIII levels reflects a
consumptive process as the plasma TAT complexes
increase as ATIII levels drop.

The level of ATIII that may be cause for concern has not
been accurately defined. However, patients who have
undergone shock and demonstrate levels of ATIII below
50–60% of normal activity have an increased morbidity and
mortality. Patients with levels below 20% have near
100% mortality.

Replacement or enhancement of ATIII concentrations
has been suggested in a number of these conditions.
 Concentrates from human sources have been available for
some time and have shown some benefits in patients with
sepsis syndrome. A recombinant form of ATIII has entered
clinical trials as a method of reducing ‘heparin resistance’ in
patients before heart surgery.

HCII is the second plasma thrombin inhibitor. The
endothelial glycosaminoglycan, dermatan sulphate has a
specific binding site for HCII. This binding site is a
hexasaccharide without structural similarity with the
pentasaccharide of heparan or heparin.

Therapies to inhibit thrombin production or
activity

Included in this category are recombinant agents equivalent
to some naturally occurring proteins and totally synthetic
agents. A most important point to note is that at times these
drug therapies will produce prothrombotic or hypercoagu-
able states. Warfarin therapy is associated with cutaneous
thrombosis. This latter effect is a result of the action of
warfarin to reduce effective protein C concentration (a
vitamin K-dependent factor) and induce a prothrombotic
protein C deficiency. Moreover, studies in patients given
warfarin immediately after myocardial revascularization
show a short period of a hypercoagulable state, due directly
to the administration of warfarin. Heparin will induce a
thrombotic state by direct or immune-mediated platelet
activation as described later.

The first group of antithrombin drugs discussed are not
direct inhibitors of thrombin but aim to slow thrombin
generation and presentation.

Vitamin K antagonists

Reduction in clotting factor activity is produced when
patients are given vitamin K antagonists. The first oral
anticoagulant used was dicoumarol that was isolated from
spoil clover. This agent had a poor absorption and non-
linear kinetics and is no longer used. The three widely used
drugs are warfarin, phenprocouman, and acenocouman.

Warfarin is the best known of this class of agent and is
used prophylactically in atrial fibrillation, venous throm-
bosis, pulmonary embolism, and in patients with prosthetic
heart valves. The effect of warfarin is monitored by the PT
or the International Normalized Ratio (INR). The INR was
developed by the World Health Organization in the early
1980s to eliminate problems in oral anticoagulant therapy
caused by variability in the sensitivity of different com-
mercial sources and different batches of thromboplastin.
The INR is derived by raising the observed ratio of PT in
control and patient plasma to the power of an International
Sensitivity Index (ISI). The ISI is a measure of the response
to a thromboplastin preparation and is typically between 2
and 2.6 for most commercial rabbit-brain thromboplastins.
The INR has for no obvious reason been only slowly
adopted within North America compared with the rest of the
international community. The PT ratio, which has been
adopted by centres in North America, is not directly
interchangeable with INR. This adds some confusion
when discussing results from studies of the effects of an
anticoagulant regimen on outcome. An INR of 2.5 is
adequate for treating venous thrombosis, pulmonary
thromboembolism and for atrial fibrillation. An INR of 3.5
is the target for patients with heart valves.

Warfarin can be given intravenously but is usually given
orally and is well absorbed. Peak plasma concentrations
of warfarin occur about 90 min after ingestion. The plasma
half-life is about 36 h and it usually takes about 3 days of
daily dosing to achieve a steady-state concentration of the
drug. A prolonged coagulation time requires more than a
25% decrease in factor activity. This takes about 8–24 h
following ingestion of warfarin. The peak effect of a single
dose occurs at 36–72 h and lasts about 5 days.

Warfarin is a racemic mixture of about equal amounts of
so-called R and S forms. The S form is about five times
more potent as a vitamin K antagonist than the R form and is
oxidized in the liver to hydroxywarfarin that is excreted in
the bile. The R form is metabolized to warfarin alcohols,
which are excreted by the kidney. About 97% of warfarin in
the circulation is bound to albumin. Given the above
confounding variables it is not surprising that the biological
effect of warfarin to prolong the PT can be significantly altered by a multitude of other therapeutic interventions as shown in Table 2.

**Table 2** Drugs that alter PT by interaction with warfarin

<table>
<thead>
<tr>
<th>Drugs that change plasma concentration of warfarin</th>
<th>Additive anticoagulant effect with no change in plasma concentration</th>
<th>Unknown mechanisms</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Prolonged prothrombin time</strong></td>
<td>Inhibit vitamin K cycle</td>
<td>Ketoconazole/fluconazole</td>
</tr>
<tr>
<td>Inhibit S isomer clearance</td>
<td>Second/third generation cephalosporins</td>
<td>Tamoxifen</td>
</tr>
<tr>
<td>Metronidazole</td>
<td>Inhibit coagulation and/or platelet function</td>
<td>Phenytoin</td>
</tr>
<tr>
<td>Sulphinpyrazone</td>
<td>Heparin</td>
<td>Erythromycin</td>
</tr>
<tr>
<td>Trimethoprim</td>
<td>Aspirin/ticlopidine/clopidagrel</td>
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<tr>
<td>Disulphiram</td>
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<td></td>
</tr>
<tr>
<td>Inhibit R isomer clearance</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cimetidine</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Omeprazole</td>
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<tr>
<td>Inhibit both S and R isomer clearance</td>
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</tr>
<tr>
<td>Amiodarone</td>
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</tr>
<tr>
<td><strong>Reduced prothrombin time</strong></td>
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<tr>
<td>Reduced absorption</td>
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<td>Penicillins</td>
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<tr>
<td>Cholestyramine</td>
<td></td>
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</tr>
<tr>
<td>Increased metabolism</td>
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<tr>
<td>Barbiturates</td>
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<tr>
<td>Rifampicin</td>
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<tr>
<td>Griseofulvin</td>
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<td></td>
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<tr>
<td>Carbamazepine</td>
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</tr>
</tbody>
</table>

**Vitamin K** is necessary for the carboxylation of glutamate residues of factors VII, X, IX, prothrombin, and protein C. Vitamin K antagonists prevent this process. In other words, the plasma concentration of these factors is normal but their function is impaired. With normal liver function, a dose of vitamin K will reverse this functional defect in about 4–6 h. However, it takes up to a week to re-establish the anticoagulant effect of warfarin after vitamin K. Fresh frozen plasma will transiently reverse the effect of warfarin although this requires volumes of about 10–15 ml kg⁻¹.

In the context of the perioperative period, there are few evidence-based criteria to guide patient management. The risks of thromboembolism if the patient is not anticoagulated must be balanced with the risks of excessive intraoperative bleeding. For example, in a survey of patients having dental surgery, the incidence of significant bleeding (0.05%) was similar to those with thrombotic complications (0.03%).⁶⁷ The major difference was that four of the five patients with thrombotic complications died as a consequence of these events. Although the likelihood of significant bleeding may be greater with more major procedures such as bowel or orthopaedic surgery, this must be balanced against the magnitude of the thrombotic risk and its effects. Although it is common practice to stop warfarin up to a week before surgery, and substitute the more readily reversible heparin, there is little current evidence to show this is an absolute necessity. This is especially the case in patients with prosthetic cardiac valves.¹⁵ One other recent review suggested that withholding warfarin for 48 h was associated with a 1 point fall in the INR from about 3.5 to 2.5 in patients with prosthetic valves. This value was not associated with increased bleeding or increased thrombotic episodes over the transient period of drug withdrawal.⁷⁹

One further area of concern is the patient taking oral anticoagulants whom becomes pregnant. In particular, the management of women with prosthetic heart valves during pregnancy poses a particular challenge, as there are no available controlled clinical trials to provide guidelines for effective antithrombotic therapy. Warfarin is teratogenic and should not be given in the first trimester of pregnancy. However, subcutaneous (s.c.) administration of heparin is reported to be ineffective in preventing thromboembolic complications. A recent literature review¹² suggested that the regimen associated with the lowest risk of valve thrombosis was the use of oral anticoagulants throughout pregnancy. Although this approach was associated with warfarin embryopathy in 6.4% (95% CI 4.6–8.9%) of live births, this was less than the 9.2% (95% CI 5.9–13.9) risk of valve thrombosis using heparin only between 6 and 12 weeks' gestation.

**Heparins**

Heparin is a naturally occurring negatively charged sulphated polysaccharide with a complex structure. It is a glycosaminoglycan formed from alternating residues of D-glucosamine and L-iduronic acid. The important part of the molecule with regard to anticoagulation is thought to be a specific, ATIII binding, pentasaccharide sequence found in about one-third of molecules of UFH, with a lower proportion in molecules of the low-molecular weight product.³² ³³ Heparin is located mostly in mast cells in lungs, intestine, and liver in mammals. Heparin was originally isolated from liver during investigations to ascertain if the phospholipid component of cephalin would
cause clotting. Since the discovery of heparin in 1916 by McLean, numerous physiological actions have been proposed for this agent. The finding of heparin-rich mast cells in tissues where the inside and outside of the body are in close proximity (the skin, lung, and gut) suggests a primary anti-inflammatory or immunological role for this agent. This concept is strengthened when we consider first, that heparin alone has no direct effects on coagulation, and secondly, is found in lower orders of the animals, such as molluscs, which lack a coagulation system.

**UFH**

Standard preparations of heparin are unfractionated (UFH), derived from either porcine intestine or bovine lung and prepared as either calcium or sodium salts. The number and sequence of the saccharides is variable, with molecular weights ranging from 3000 to 30 000 Da, with a mean of 15 000 Da representing 40–50 saccharides in length. There is no apparent difference between any of the available forms of UFH with respect to their pharmacology or anticoagulant profile.

**Low-molecular weight heparin**

Low-molecular weight heparins (LMWH) are produced from UFH by chemical or enzymatic depolymerization. This produces marked changes in the properties of the heparin and leads to the difference between the clinical effects, pharmacokinetics and pharmacodynamics of LMWH compared with UFH. LMWHs have mean molecular weights of 4000–6500 Da, although the range is 2000–10 000 Da. There are significant variations between the different commercial preparations according to the method used in their production as shown in Table 3.

**Mechanism of increased anticoagulation with heparin**

The binding of heparin to ATIII is highly specific, reversible, and does not inactivate the heparin molecule. ATIII has an intrinsic low level of activity, mediated by an arginine centre that binds to activate serine proteases of the coagulation cascade. Binding with heparin dramatically increases this inhibitory effect. The association constant between thrombin and the ATIII-heparin complex is increased 10 000-fold and the rate constant for the reaction increased some 2000-fold. This action reduces the half time of inhibition of thrombin in plasma to 10 ms.

The augmentation of the inhibitory effect of ATIII comes about in three separate ways. First, heparin attaches to a small, high affinity site on ATIII to produce a conformational change at the reactive site (Fig. 8). This occurs with
the pentasaccharide sequence alone and results in a 100-fold increase in inhibition of not only thrombin but also factor Xa and certain other proteases, including coagulation factors XIIa, XIa, IXa, plasmin, and kallikrein. Interestingly, loss of affinity for the pentasaccharide at this site (such as found in ATIII Geneva or ATIII Rouen that have point mutations of this site) leads to only a mild tendency to develop venous thrombosis, suggesting other effects of heparin on ATIII.

The second effect of heparin is via a larger, but low affinity, site that extends from the pentasaccharide site to the pole of the ATIII molecule near the active centre. The active centre carries a positive charge, which tends to repel proteases with a positive charge at the active centre such as thrombin. Addition of the negative charge carried on the heparin will neutralize this effect (Fig. 8). This may explain the observation that the addition of heparins with a longer chain length (thus more available negative charges) has a much greater effect to inhibit thrombin than it does to inhibit plasmin or factor Xa. Increased negativity will also not affect proteases with a neutral active site such as plasmin, or to a lesser extent factor Xa.

The third effect is also specific to the inhibition of thrombin, which has charged exosites away from the active centre. One of these exosites attaches to longer (more than 18 residues) non-specific parts of the heparin molecule which are bound to the high affinity site by their pentasaccharide sequence. This long tail ensures a certain afﬁnity, site that extends from the pentasaccharide site to the pole of the ATIII molecule near the active centre. The active centre carries a positive charge, which tends to repel proteases with a positive charge at the active centre such as thrombin. Addition of the negative charge carried on the heparin will neutralize this effect (Fig. 8). This may explain the observation that the addition of heparins with a longer chain length (thus more available negative charges) has a much greater effect to inhibit thrombin than it does to inhibit plasmin or factor Xa. Increased negativity will also not affect proteases with a neutral active site such as plasmin, or to a lesser extent factor Xa.

The importance of chain length in relation to the range and speciﬁcity of the catalytic action of heparin can be best appreciated from studies of small, semi-synthetic oligosaccharides. These studies showed that with less than 18 saccharides, there was little activity to inhibit thrombin (as shown using the thrombin time coagulation test) compared with a 140-fold inhibition of this test with molecules with more than 18 residues. Increased factor Xa inhibition was detected with both chain lengths but was 2–3-fold more potent with lower numbers.

Following the reaction between the active site of the protease and the reactive site of ATIII, a further conformational change in ATIII occurs that causes it to envelop the protease. This change also reduces the afﬁnity of ATIII for heparin, which is released to participate in further ATIII-protease reactions. Heparin has two further antithrombotic actions in addition to those mediated via ATIII.

First, heparin can activate the other major circulating antithrombin, HCII. This activation does not require the pentasaccharide sequence but does require heparins of greater than 7200 Da or 24 saccharide units in length. Activation of HCII is assumed to be a non-specific effect of heparin related to the total charge on the molecule rather than acting via a specific receptor. This non-specific activation may explain why the HCII-dependent effect requires a 10-fold higher concentration (typically >4 IU ml⁻¹) than that required to activate ATIII. Secondly, heparin will stimulate the release of TFPI, reducing prothrombinase production via the extrinsic pathway. Plasma concentrations of TFPI increase 2–6-fold following heparin injection. This increase occurs with UFH and LMWH.

Pharmacokinetics and pharmacodynamics of heparins

Similar to the mechanism of action of UFH and LMWH, the pharmacokinetics and dynamics have a number of differences and some similarities. In particular, neither signiﬁcantly cross the placenta. Moreover, the plasma concentrations of heparin are not uniformly related to the anticoagulant effect produced and there is a wide variability in dose–response effects in patients.

UFH

The pharmacokinetics of UFH are complex. Heparins are poorly absorbed from the gastrointestinal tract and can cause haematomas after intramuscular injection. They are therefore usually administered by s.c. or intravenous (i.v.) injection. I.v. injection is the preferred route when a rapid anticoagulant effect is needed. However, similar levels of anticoagulation can be achieved, with onset delayed by 1 or 2 h, by the s.c. route, if sufﬁcient doses are used. Studies suggest the safety of the two routes is comparable.

The heterogeneity of heparin molecules produces great variability in the plasma concentration of the agent in relation to the dose administered. A three-compartment model best describes the kinetics of UFH in humans. After injection, plasma levels initially decline rapidly a result of redistribution and uptake by endothelial cells. More than 50% of heparin circulates bound to proteins including platelet factor 4, histidine-rich glycoprotein, vitronectin, ﬁbronectin, and vWF. The first three of these also reduce its bioavailability and activity. Raised concentrations of these proteins may account for the heparin resistance seen in malignancy and inﬂammatory disorders. The release of platelet factor 4 from activated platelets may reduce heparin concentration at the site of clot formation, contributing to the reduced efficacy of heparin against clot-bound thrombin.

Heparin clearance is non-linear and elimination occurs by two separate processes; a rapid mechanism, which is readily saturated at clinically therapeutic concentrations, and a slower process involving first-order kinetics. The rapid, saturable phase of heparin clearance is thought to be a result
of cellular degradation by macrophages, which internalize the heparin, then depolymerize and desulphate it. Saturation occurs when all the receptors have been utilized and further clearance depends on new receptor synthesis. This process explains the poor bioavailability of heparin after low-dose s.c. injection: the slow rate of absorption barely exceeding the capacity for cellular degradation. Significant plasma levels can only be achieved by saturation of these receptors with a loading dose. The slower phase of heparin elimination is a result of renal excretion. This complex mechanism of elimination means that as the dose of heparin is increased, the elimination half-life appears to increase in duration. A bolus of 25 unit kg\(^{-1}\) has an apparent half-life of 30 min which increases to 60 min with a dose of 100 unit kg\(^{-1}\) and this half-life duration is further increased to 150 min when the bolus dose is 400 unit kg\(^{-1}\).\(^{6,17}\) Surprisingly, no consistent report of the effects of renal or hepatic dysfunction on the pharmacokinetics of heparin have been described.\(^{29,31}\)

**LMWH**

The affinity of plasma proteins for LMWHs is much less than for UFH, so that only 10% is protein bound. Moreover, LMWHs are not subject to the rapid degradation that UFH suffers as they are not inactivated by platelet factor 4 and do not bind to endothelial cells or macrophages. This produces nearly complete bioavailability, compared with 40% for low-dose s.c. UFH, and this guarantees a more predictable anticoagulant action. They are almost completely absorbed following s.c. injection.

In contrast to UFH, the LMWHs exhibit linear pharmacokinetics with proportionality between anti-Xa (and anti-IIa in some cases) plasma concentration and dose. Their distribution volume is close to the blood volume. Similar to UFH they are partially metabolized by desulphation and depolymerization.

Although the clearance of LMWH is dependent on renal excretion, producing a half-life two to four times as long as UFH, this is not clinically relevant until there is severe renal disease and creatinine clearance values less than 15 ml min\(^{-1}\) are achieved.\(^{35}\)

Urinary excretion of anti-Xa activity for enoxaparin, dalteparin, and nadroparin, all given at doses for prevention of venous thrombosis, is between 3 and 10% of the injected dose. However, these LMWHs differ in the extent of their non-renal clearance, resulting in different apparent elimination half-life values and relative apparent bioavailability.

The LMWHs available differ in the distribution of molecular weights, *in vitro* potency, bioavailability, anti-Xa activity,\(^{22}\) and consequently their pharmacodynamic behaviour, recommended dose regimen, and efficacy/safety ratio.\(^{29}\) Because of these differences among LMWHs, the clinical profile of a given LMWH cannot be extrapolated to another one or generalized to the whole LMWH family. Table 4 shows data for potency, plasma half-life, and bioavailability for certain currently available agents.

### Table 4 Potency, plasma half-life and bioavailability of various LMWHs compared with UFH

<table>
<thead>
<tr>
<th>Preparation</th>
<th>Anti-Xa to -IIa ratio</th>
<th>Half-life (min)</th>
<th>Bioavailability (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>UFH</td>
<td>1</td>
<td>30–150</td>
<td>10–20</td>
</tr>
<tr>
<td>Nadroparin</td>
<td>3.6:1</td>
<td>132–162</td>
<td>89</td>
</tr>
<tr>
<td>Enoxaparin</td>
<td>3.8:1</td>
<td>129–180</td>
<td>91</td>
</tr>
<tr>
<td>Dalteparin</td>
<td>2.7:1</td>
<td>119–139</td>
<td>87</td>
</tr>
<tr>
<td>Tinzaparin</td>
<td>1.9:1</td>
<td>111</td>
<td>90</td>
</tr>
<tr>
<td>Certoparin</td>
<td>2.0:1</td>
<td>240</td>
<td>90</td>
</tr>
<tr>
<td>Ardeparin</td>
<td>1.9:1</td>
<td>200</td>
<td>90</td>
</tr>
<tr>
<td>Reviparin</td>
<td>3.6:1</td>
<td>180</td>
<td>90</td>
</tr>
</tbody>
</table>

### Monitoring heparin effects

Monitoring is routinely performed during therapy with UFH for a number of reasons.

There is a marked variation in the initial anticoagulant response to a fixed dose of UFH.

Variability in saturation of cellular binding and degradation will enhance the non-linear response following repeat dosage.

The risk of recurrent thromboembolism is reduced if the effect of heparin is maintained above the lower therapeutic limit.\(^{37,48}\)

Direct measurement of heparin concentration is not possible, although protamine titration can be used to ascertain blood levels. The commonly used laboratory test of heparin function is the aPTT, which measures the effect of heparin-activated coagulation inhibition. The therapeutic range most commonly quoted is an aPTT between 1.5 and 2.5 times the control value. However the commercially available kits for measurement of aPTT differ in their sensitivity to heparins. This suggests that the protamine titration method of monitoring may be more robust.\(^9\)

The higher bioavailability of LMWH and their longer half-life produce a more predictable anticoagulant response. These properties allow once daily s.c. administration for prophylaxis of deep venous thrombosis.\(^{33,37}\) Although no laboratory monitoring is required for the majority of patients receiving LMWH, it is important to check anti-Xa activity in those who are renally impaired, as pregnancy progresses (with associated fluctuations in weight and plasma volume), and where thrombosis and/or haemorrhage occur or are suspected while on treatment. The anti-Xa assay used must also be calibrated against the brand of LMWH prescribed.

### Side effects of heparin

Although haemorrhage is rare with prophylactic doses of either UFH or LMWH given alone, it is a frequent complication of therapeutic heparin administration. The greater the dose of heparin and therefore the greater its anticoagulant effect, the greater the risk of haemorrhage. When comparable doses are used, the risks are similar using either the continuous i.v. or s.c. route of administration.\(^{29,51}\) Many patient factors are known to increase the risk of haemorrhage including the duration of treatment, presence
of cardiac, hepatic or renal dysfunction, aspirin or other platelet active therapy, recent surgery, and trauma or invasive procedures. There is, for example, evidence for increased, and more prolonged bleeding after emergency cardiac surgery in patients who have received platelet-active agents such as abciximab, clopidogrel, or ticlopidine, but not with agents such as eptifibatide which have a shorter duration of action.

Heparin will also impair platelet aggregation and inhibits platelet function by direct binding to platelets. It is the higher-molecular weight heparin molecules with the lower affinity for ATIII that interfere most with platelet function. These actions may be responsible for heparin-induced haemorrhage by a mechanism which is separate to its anticoagulant actions.

Approximately 30% of patients who suffer anticoagulant-related haemorrhage are found to have previously undiagnosed predisposing lesions, particularly of the gastrointestinal and genitourinary tracts. A review of this complication estimated the daily frequencies of fatal, major, and all types of haemorrhage in patients receiving therapeutic anticoagulation as 0.05, 0.8, and 2.0%, respectively, approximately twice the level expected in the absence of anticoagulation. The incidence of increased perioperative haemorrhage, which contributed to adverse outcome, has been reported variously in between 2% and 10% of patients receiving prophylactic heparin therapy.

Studies of LMWHs given for thromboprophylaxis suggest they cause an increase in wound haematomas but no change in the incidence of haemorrhage. In contrast, a significant reduction in major haemorrhage is seen when LMWHs are used to treat established thrombosis.

Although chronic heparin administration has been associated with osteoporosis and hypoaldosteronism, these remain medical curiosities compared with the relatively common and potentially life-threatening problem of heparin-induced thrombocytopenia (HIT) and especially that associated with osteoporosis and hypoaldosteronism, these remain medical curiosities compared with the relatively common and potentially life-threatening problem of heparin-induced thrombocytopenia (HIT) and especially that associated with osteoporosis and hypoaldosteronism, these remain medical curiosities compared with the relatively common and potentially life-threatening problem of heparin-induced thrombocytopenia (HIT) and especially that associated with osteoporosis and hypoaldosteronism, these remain medical curiosities compared with the relatively common and potentially life-threatening problem of heparin-induced thrombocytopenia (HIT) and especially that associated with osteoporosis and hypoaldosteronism, 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associated with osteoporosis and hypoaldosteronism, these remain medical curiosities compared with the relatively common and potentially life-threatening problem of heparin-induced thrombocytopenia (HIT) and especially that associated with osteoporosis and hyp
Two other therapies are available and licensed for use to provide anticoagulation in patients with HIT. The first is danaparoid sodium, which is a mixture of the endothelial cell glycosaminoglycans heparan, dermatan, and chondroitin sulphates. Effect is monitored using anti-Xa activity. The second agent is hirudin (Lepirudin) manufactured using recombinant technology. Use and monitoring of Lepirudin is discussed later.

Reversal of action: antidotes to heparins
The effects of UFH wear off so rapidly that an antagonist is rarely required, except after the high doses administered to facilitate cardiopulmonary bypass. Protamine is a basic protein extracted from fish sperm that combines with heparin to form a stable, inactive complex. Conventional wisdom was that equimolar amounts of protamine sulphate (1 mg protamine for 100 units of heparin) should be used to provide optimal neutralization. However, the use of protamine titration using various techniques has been shown to considerably reduce blood loss and transfusions after heart surgery using considerably lower doses of protamine. Although protamine will reduce the anticoagulant effects of heparin, it does not affect various other actions induced by heparin administration. Of relevance is the effect of heparin to increase plasma concentrations of free drug, such as diazepam and propanolol, is not inhibited or prevented by protamine administration.

With LMWHs, protamine is able to neutralize the anti-IIa but not the anti-Xa action of heparin, because of inability to bind the smaller heparin molecules. An analogous situation occurs with the use of heparinase. This enzyme will break down the larger molecular weight UFH, but this will produce lower molecular weight compounds. This may be one reason for an increase in blood loss in patients who had heparin reversal with heparinase rather than with protamine. This effect may be extremely important in relation to local anaesthetic block.

Local anaesthesia in the patient receiving heparin
Therapeutic anticoagulation is a contraindication to central nerve block unless the coagulation profile is corrected to normal. The risks associated with epidural or spinal anaesthesia in patients receiving heparin prophylaxis is a controversial subject which has been the subject of a number of recent reviews.

Reduced efficacy of heparin therapy
Although bound, thrombin can be inactivated by the heparin–ATIII complex, much higher concentrations are required than are needed to inactivate free thrombin. In addition, platelets secrete platelet factor 4, which neutralizes heparin. Clinically, this is seen as a requirement for much higher levels of heparin to prevent the extension of venous thrombosis compared with those required to prevent initiation of thrombosis. This poor efficacy of heparin is, in part, due to the fact that naturally occurring polypeptide thrombin inhibitors, such as ATIII, cannot inhibit bound thrombin.

Other agents used to inhibit thrombin
The inability of polypeptide inhibitors to directly target bound thrombin has been one reason for efforts to develop low-molecular weight, direct-acting, or site-directed, thrombin inhibitors. A further recurrent problem with the use of heparin and vitamin K antagonists is multiple sites of action. This leads to difficulties of accurate dosing and monitoring to maximize the therapeutic window and minimize the risk of bleeding. Finally, and as with nearly all of the drugs which effect the haemostatic or coagulation systems, the drive to produce newer, more specific agents, has largely come from studies in patients having interventional cardiology such as angioplasty or stent insertion. These agents are also of interest as they provide means to establish anticoagulation in the patient who has a history of type II heparin-induced thrombocytopenia and thrombocytopenia.

Hirudin
Hirudin is a polypeptide originally obtained from the medicinal leech. The molecule is now manufactured by recombinant technology with the generic name of Lepirudin. Two recombinant hirudin preparations, revasc (Novartis) and refudan (Aventis), are available for postsurgical DVT prophylaxis and alternate anticoagulant use in patients with heparin-induced thrombocytopenia. A synthetic antithrombin agent based on the combined structures of hirudin and antithrombin peptides, hirulog (Bivalirudin), is undergoing clinical trials in cardiovascular indications. Additional studies on the hirudins are being carried out to test their efficacy as anticoagulant replacements for heparin for both surgical and interventional cardiology indications. Hirudin acts by irreversible binding to the active site of thrombin. The irreversibility is a result in part to the molecule having a long carboxyl tail which binds to the anion binding exosite of thrombin (Fig. 9).

Therapeutic concentrations for r-hirudin are about 1 μg ml⁻¹ for vascular surgery and 2–4 μg ml⁻¹ for surgery with extracorporeal circulation. For concentrations up to 1 μg ml⁻¹ there is a relatively steep dose–response curve with the aPTT and this test can be used to assess anticoagulant activity. Therapeutic concentrations are adequate with an aPTT of about twice control values. Above 1 μg ml⁻¹ the
dose–response for the aPTT is almost horizontal and the test fails to accurately reflect plasma concentrations. In this circumstance, a second activator called ecarin has been shown to produce linear dose–response characteristics.63 64 Ecarin is a snake venom which directly cleaves prothrombin to produce a thrombin-like material which is inhibited by hirudin. The ecarin time test is not routine in most haematology laboratories and is expensive.

The major clinical disadvantage of hirudin preparations is their mode of excretion by renal clearance leading to a plasma half-life of about 14 h. This duration is not shortened by dialysis or haemofiltration. Significant bleeding has been reported in patients having continuous venovenous haemofiltration and hirudin infusion,47 and also in patients having cardiac surgery.51 Unlike heparin or the vitamin K antagonists, there is no clinically proven antagonist for hirudin. Studies in animals have shown partial benefits to reduce bleeding using either activated prothrombin complex or recombinant factor VII in one report,55 or factor VIII and DDAVP in another.11 This is likely to prove one of the limiting factors for the optimal development of hirudin. One other problem is that patients have been reported to develop antibodies to the recombinant form,56 which have been associated with hypersensitivity reactions.

**LMWH inhibitors**

**Arginine analogues**

A number of chlormethyketones have been used in laboratory-based medicine as direct inhibitors of thrombin; however, they are too toxic for use in humans. Arginine is the amino acid residue at the active site of the serine protease inhibitors such as ATIII. A combination of these chemistries is found in TAME (tosyl-arginine methyl ester) which has formed the basis of two direct-acting inhibitors, argatroban and napsagatran.69 These small molecules are able to fit easily into the active pocket of thrombin and other serine protease and bind with great affinity (Fig. 9). Napsagatran has reached phase II studies in humans.

**Argatroban** has been extensively investigated and is the first clinically approved antithrombin agent. The molecular properties of argatroban (small, fast, selective, with reversible inhibition of the thrombin catalytic site, and similar affinity for inhibiting both clot-bound and soluble thrombin) offer the potential for significant antithrombotic efficacy with minimal systemic anticoagulant effects. The i.v. agent, Novastan (a brand of argatroban), is currently approved for clinical use in Japan for the treatment of peripheral arterial occlusive disease, and in the USA for anticoagulation in patients with HIT and thrombosis. Novastan is in advanced clinical development in other countries for several indications, including therapy in heparin-induced thrombocytopenia and thrombosis syndrome, and as adjunctive therapy to thrombolytic agents in acute myocardial infarction.

The pharmacokinetic profile of argatroban is described by a two-compartment model with first-order elimination; mean (sd) clearance, steady-state volume of distribution, and half-life values from 40 healthy volunteers were 4.7 (1.1) ml min⁻¹ kg⁻¹, 179.5 (33.0) ml kg⁻¹, and 46.2 (10.2) min, respectively. Clearance is about 20% lower in the elderly. Kinetic analysis shows no significant differences in these variables in patients with renal dysfunction. With hepatic impairment, the maximum concentration and half-life of argatroban were increased approximately 2- to 3-fold, associated with a reduction in clearance to 25% of that in healthy volunteers.74 Control of anticoagulation is by the aPTT or ecarin clotting time. aPTT or ACT and plasma argatroban concentrations were well correlated. Dose regimens vary based on the clotting time but are typically in the range of 0.5–4 μg kg⁻¹ min⁻¹, as a continuous infusion for anticoagulation during haemofiltration or use of intra-aortic balloon counterpulsation, and about 2 μg kg⁻¹ min⁻¹ after a bolus of 100 μg kg⁻¹ for abdominal aortic surgery.58 These doses produce an ACT of about 180–200 s.

Argatroban also appears to have certain other interesting effects that may increase its use and broaden its therapeutic indications. The first relates to the possibility that there is an increase in nitric oxide production associated with infusion of argatroban, which may prove useful in providing arteriolar dilatation in peripheral vascular disease.77 This nitric oxide effect may be a result of the presence of the arginine moiety in the molecule. The second aspect is related to effects on the cerebral circulation. A number of recent semi-anecdotal studies have suggested that ischaemic cerebral tissue can be revascularized and blood flow increased in humans during infusion of argatroban.41 43

Several of the synthetic thrombin inhibitors such as melagatran55 are also being developed for oral use.70 As the therapeutic index of thrombin inhibitors is narrower than that of heparin, this route may not be an optimal approach for the development of these agents. However, there is a potentially huge market for this class of compound, which could replace drugs such as heparin and warfarin in the next few years. Despite several unresolved developmental issues, the thrombin inhibitors provide an alternative to heparin anticoagulation and may prove to be useful in clinical use.

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