Haematology

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Thrombophilia

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Venous thromboembolism (VTE) has an annual incidence of around one per 1,000 of the UK population. It is exceptionally rare in childhood but becomes more common with increasing age, reaching one in 100 in the elderly. Fatality rates from pulmonary embolism vary between 1% in the young and 10% in older subjects with comorbid conditions. There is significant long-term morbidity due to chronic post-phlebitic symptoms which ensue in a third of cases of deep vein thrombosis (DVT).

DVT results from the interaction of inherited and environmental factors. Of Virchow's triad, stasis and hypercoagulability predominate in the deep veins rather than vessel injury. Examples of inherited hypercoagulability are deficiency of a physiological anticoagulant (eg antithrombin) or mutation in a gene resulting in inefficiency of a physiological anticoagulant mechanism (eg factor V Leiden). Acquired hypercoagulability includes changes induced by pregnancy and oral contraception.

'Thrombophilia' describes an inherited predisposition of a subject who is at increased risk of VTE. However, this usage is too restrictive. For example, an individual who suffers a DVT when the majority do not, such as postpartum, could be deemed to be thrombophilic even if no heritable condition is detectable using current methods. It is perhaps more useful to develop the concept of a continuum of risk of thrombosis dependent upon the interaction of inheritance with the additional, sometimes modifiable, acquired risk factors. The heritable thrombophilias, diseases associated with increased thrombotic risk, and additional risk factors are listed in Tables 1–3, respectively.

Many prothrombotic states which result from heritable and acquired thrombophilia are due to interference in key anticoagulant mechanisms centred around the generation of thrombin and factor Xa during blood coagulation. These are illustrated in Fig 1.

Heritable thrombophilia

Interest in VTE as a heritable condition gained momentum in 1994 with the recognition that a common polymorphism in the gene for factor V (factor V Leiden) is associated with resistance to the anticoagulant action of protein C (protein C resistance) and is a procoagulant state (Fig 1). Heterozygotes (5% of the UK population, up to 10% in some northern European countries) carry a lifelong risk of venous thrombosis about fourfold greater than non-carriers. The risk is higher still in homozygotes. Around 20–40% of patients with venous thrombosis in the UK are heterozygous. Frequency depends on the selection criteria used for testing: for example, presence of family history of VTE and apparent spontaneity of the acute episode, both of which increase the frequency of factor V Leiden detection.

Prior to discovery of inherited protein C resistance, diagnosis of heritable thrombophilia was unusual as only deficiencies of the physiological anticoagulants antithrombin, protein C and protein S were recognised. These are found in only 2–10% of subjects with VTE. Another point mutation, in the gene for prothrombin, has recently been shown to carry an increased level of risk. Prothrombin G20210A, associated with increased plasma levels of prothrombin, has a prevalence of around 1% in the UK. Up to 10% of selected subjects with DVT are heterozygous for this gene. Factor V Leiden and prothrombin G20210A are rare in Asians and Africans.

The heritability of thrombophilia also depends on genes that determine plasma concentrations of factors VIII, IX and XI and fibrinogen. The risk of venous

Table 1. Thrombophilic states with a heritable component.

Activated protein C resistance/ Factor V Leiden	Raised factor VIII concentration
Prothrombin G20210A	Raised factor IX concentration
Antithrombin deficiency	Raised factor XI concentration
Protein C deficiency	Raised fibrinogen concentration
Protein S deficiency	Hyperhomocysteinaemia

Table 2. Systemic diseases and conditions associated with increased risk of venous thrombosis.

- Obesity
- Cancer
- Haematological diseases:
  - haematological solid tumours
  - myeloproliferative disease (essential thrombocythaemia, polycythaemia rubra vera)
  - paroxysmal nocturnal haemoglobinuria
  - sickle cell disease
- Acute myocardial infarction
- Acute stroke
- Systemic lupus erythematosus
- Antiphospholipid syndrome
- Behçet's syndrome
- Inflammatory bowel disease
- Nephrotic syndrome
- Limb paralysis
- Postoperative state

thrombosis rises with plasma concentrations of each of these. However, environmental factors also determine these levels: for example, factor VIII and fibrinogen are acute-phase proteins.

Finally, increased plasma concentration of the amino acid homocysteine is determined in part by genetic variation in the enzymes responsible for its synthesis, cystathione synthetase and methylenetetrahydrofolate reductase (MTHFR) and also by the levels of vitamins B12, B6 and folic acid. Hyperhomocysteinaemia is accompanied by increased risk of both venous and arterial thrombosis. Although a common mutation in the gene for MTHFR associated with higher levels of homocysteine is recognised, the mutation alone does not convey increased risk of venous thrombosis, suggesting other factors may be more important, such as folate status.

Factor V Leiden and prothrombin G20210A are quite prevalent so co-inheritance with other thrombophilias is not rare. Risk of thrombosis is increased further in such cases, for example when factor V Leiden and protein C deficiency occur together.

Pregnancy failure has also been reported to be more common in women with heritable thrombophilia. The significance and therapeutic consequences of this observation remain to be determined.

Table 3. Additional risk factors for venous thromboembolism.

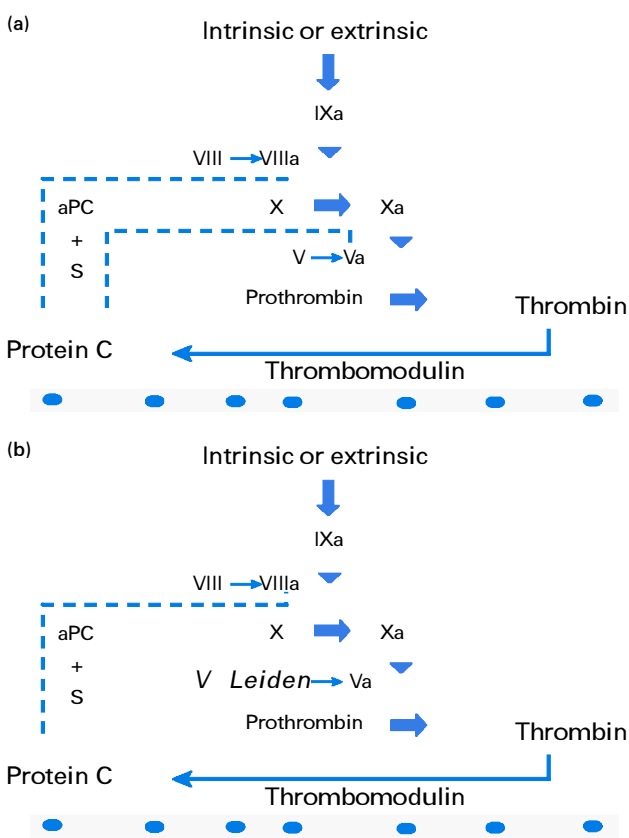
- Increasing age
- Hormone use
  - oral contraception
  - hormone replacement therapy
- Pregnancy and the puerperium
- Immobility
  - due to illness
  - due to splinting
  - during travel
- Medications
  - chemotherapy
  - tamoxifen
  - heparin (in heparin-induced thrombocytopenia
- Indwelling venous catheters and devices

The pathogenetic mechanisms of arterial thromboembolism differ from those in the venous system, vessel wall injury being a major factor. A contribution by factor V Leiden to risk of coronary artery and cerebrovascular occlusion has been sought, but it is of minor, if any, importance with the possible exception of childhood stroke. It is noteworthy that longevity is not adversely influenced by inheritance of factor V Leiden.

Acquired risk factors

Subjects with heritable thrombophilia do not have thrombosis most of the time. A trigger is usually necessary. Increasing age makes a significant contribution through as yet unrecognised mechanisms. Other contributory events are listed in Table 3. Use of the combined oral contraceptive (OC) is worthy of emphasis as its interaction with heterozygosity for factor V Leiden is multiplicative rather than additive. The increased risk of thrombosis of around fourfold associated with OC use rises to 30–40-fold in users with factor V Leiden. Despite this, most such women will not develop thrombosis. One mechanism of hypercoagulability from combined OC use is induction of protein C resistance similar to that associated with factor V

Fig 1. Some key interactions of physiological anticoagulants. (a) Thrombin, generated through the coagulation cascade, is diverted to an anticoagulant action via interaction with thrombomodulin on endothelial cells. Thrombin/thrombomodulin activates protein C which, with its cofactor protein S, inactivates the activated forms of the coagulation factors V and VIII. In this way, thrombin inhibits its own generation to prevent unchecked clot formation. Thrombin is also controlled by antithrombin (not shown), the inhibitory action of which is massively enhanced by heparans present on the endothelium.
(b) The gene for factor V Leiden has a point mutation which leads to generation of a form of activated factor V which is insensitive to inhibition by activated protein C/protein S. Thrombin generation is less closely checked as a result, giving rise to a prothrombotic state (aPC = activated protein C; S = protein S).



Leiden. Both this effect and thrombosis risk are greater with third-generation than with second-generation products which differ principally in the progestagen components.

There has been considerable recent interest in VTE associated with long haul air travel, but the frequency of events has not been determined and is likely to have been overestimated. The risk in subjects with heritable and acquired thrombophilias has not yet been established.

Systemic diseases contribute significantly to the occurrence of thrombosis, notably antiphospholipid syndrome and cancer, including occult tumours, because of high prevalence and an especially high risk of recurrent thrombosis. In antiphospholipid syndrome the presence of persistent antiphospholipid antibody is accompanied by thrombosis and/or recurrent pregnancy failure. Other associations include thrombocytopenia and livedo reticularis. In contrast to heritable thrombophilia, arterial occlusive events are also common, especially stroke. Diagnosis requires demonstration of anticardiolipin antibody or lupus anticoagulant. In some cases both antibodies are present.

Diagnosis of thrombophilia

Laboratory tests for heritable thrombophilia are readily available, but their clinical value is far from clear. Particular issues arise in genetic testing for a generally late onset disorder with low fatality rate and incomplete penetrance. In investigating for thrombophilia it is essential to consider what can be achieved and whether harm can result.

Thus, thrombosis recurrence rates are no higher in subjects with DVT who carry factor V Leiden and there is no difference in response to anticoagulant therapy. The same is probably true for the other heritable thrombophilias, although insufficient information is available for antithrombin deficiency and combined defects where recurrence rates may be higher. In the current state of knowledge, diagnosis of heritable thrombophilia does not affect treatment of an acute episode.

The likely impact of the increased knowledge of heritable thrombophilias is in prevention of VTE in clinically unaffected carriers, but this has yet to be realised. Screening of populations at risk has been advocated but its effectiveness is open to doubt. For example, although there is a multiplicative effect between factor V Leiden and OC use, the background rate of venous thrombosis in young fit women is low. Thus, several million women must be screened to prevent a single fatal event. Furthermore, some women would avoid OC as a result of testing, with the inevitable increased risk of pregnancy. The situation is similar in screening women prior to the use of hormone replacement therapy, although the background rate of thrombosis is higher in this older population.

Case finding to identify relatives at increased risk is likely to be a more effective approach to thrombosis prevention. However, most heterozygotes will never suffer venous thrombosis and the psychological and lifestyle risks associated with diagnosis of a genetic predisposition should not be underestimated. There is also a risk of diagnostic error, particularly in relation to deficiencies of proteins C and S, where there is overlap between levels in heterozygotes and unaffected individuals. Testing for heritable thrombophilia should not be used indiscriminately and counselling must be available before and after testing.

Benefit is likely (although it has not been demonstrated conclusively) only from testing in families where there is a history of venous thrombosis in more than one individual, especially at a relatively young age and apparently unprovoked.

There is no reason to test adults with arterial thrombosis for heritable thrombophilia. Factor V Leiden and anticoagulant deficiencies do not make a significant contribution to pathogenesis. Raised fibrinogen and homocysteine are both associated with arterial occlusive events, but their detection does not influence therapy at present. Trials of folate supplementation as a preventive measure in arterial disease are underway.

The value of testing for antiphospholipid antibodies in subjects with thrombosis is more obvious because of the high rates of recurrence, indicating a need for long-term oral anticoagulant therapy in some cases. Also, the effectiveness of antithrombotic therapy in improvement of pregnancy outcomes in women with antiphospholipid syndrome and recurrent pregnancy failure has been demonstrated in a randomised clinical trial.

Key Points

- The pathogenesis of venous thromboembolism (VTE) is multifactorial
- Both inherited and acquired factors contribute
- Thrombophilic genotypes are common in the general population
- Many individuals with factor V Leiden and prothrombin G20210A will remain asymptomatic
- Laboratory tests for heritable thrombophilia are not indicated as a routine in all subjects with VTE
- Consideration should be given to the benefits and potential harm of genetic testing before embarking upon family studies for heritable thrombophilia
- Population screening is currently not indicated

Conclusions

There have been major advances in our understanding of the pathogenesis of VTE which have gone a long way towards

explaining the occurrence of thrombosis in an individual. The contribution of this knowledge to better management and prevention of venous thrombosis has not yet been defined.

Further reading

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Inherited coagulation disorders

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Physiology

Injury to a blood vessel initiates a series of events which result in controlled haemostasis. This involves adhesion and aggregation of platelets and activation of the clotting cascade to form a fibrin clot.

Platelets

When the vascular endothelium is disrupted, platelets adhere to exposed collagen. Although platelets have collagen receptors, under conditions of high shear this interaction is mediated via von Willebrand factor (VWF) which binds to the collagen and also to glycoprotein (GP) Ib on the platelet surface. Platelets then form a plug by binding to each other, a process known as aggregation. They are able to do this because when they are activated a fibrinogen receptor on their surface (GP IIb/IIIa) becomes able to bind fibrinogen. This symmetrical molecule can bridge the gap between platelets, holding them together.

Key Points

Platelet adhesion and aggregation, and blood coagulation are all necessary for haemostasis

A careful history and the judicious use of screening tests are necessary to diagnose a bleeding disorder

Haemophilia is an X-linked deficiency of factor VIII or IX in which clinical severity is predicted by factor levels

von Willebrand disease is a common, but complex, bleeding disorder which is underdiagnosed

Platelets contain storage granules and, when activated, they release their contents which recruit and activate more platelets. Inherited defects include:

- inherited thrombocytopenia
- GP Ib deficiency (Bernard-Soulier disease)
- GP IIb/IIIa deficiency (Glanzmann's thrombasthenia)
- a lack of storage granules (storage pool disease).

These are rare diseases and will not be discussed further.

Coagulation

The classic coagulation cascade is represented in diagrammatic form in Fig 1. It is divided into the intrinsic pathway which begins with contact activation, and the extrinsic pathway initiated by the tissue factor/factor VII (TF/VII) complex. Both these pathways result in activation of factor X which then cleaves prothrombin (II) to release thrombin. This simplified version of coagulation is most useful in interpreting coagulation screen results as these pathways are followed in the test tube. However, physiologically, the TF pathway is important, and *in vivo* coagulation is initiated when perturbed endothelial cells express TF on their surface. Indeed, complete deficiency of the contact factors has no effect on haemostasis. The TF/VII complex, in addition to directly activating factor X, also activates factor IX, and *in vivo* coagulation proceeds this way (Fig 2). Factor IXa in conjunction with its cofactor, factor VIII, then activates factor X. Factor VIII deficiency (haemophilia A) and factor IX deficiency (haemophilia B) are the most important coagulation disorders. Patients with factor XI deficiency exhibit a variable bleeding disorder. It seems that thrombin can activate factor XI, which can then activate factor IX in a positive feedback loop which is important for maintaining coagulation.

The diagnosis of a bleeding disorder

The history is important. The bleeding pattern may give a clue as to whether it is