

This may be the very first illustration of a deep venous thrombosis at autopsy. It comes from 'An essay on the proximate cause of the disease called Phlegmasia Dolens' by David D Davis in *Med.Chir.Trans. Lond.* 1823, 12, 419-60. I found it in the Aberdeen Medical School

Library when I was writing up my thesis in 1973.

It is the case of Miss L who had had a swollen and painful leg during life. In fact, the clinical picture of deep venous thrombosis had been recognised much earlier, and even the forerunner of the so-called economy class syndrome. For example, in 1810 Ferriar described a patient 'fatigued by a journey into the country, and lying in a bed afterwards not perfectly aired' (*Medical Histories and Reflections*, Volume iii, Cadell and Davies, London). Charles Warlow

WHAT NEUROLOGISTS NEED TO KNOW ABOUT OUTSIDE NEUROLOGY

Thrombophilia

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'I call him a perfect physician who judges it better to abstain from treatment than pursue one which might perturb the course of the malady' (Maimonedes, 1125-1204). This might well apply to long-term anticoagulation, with the inevitable risk of bleeding, in a patient with deep venous thrombosis who is found to have factor V Leiden. There is no evidence that the risk of recurrence is any higher than in a comparable patient with no evidence of inherited thrombophilia.

Table 1 Systemic diseases and conditions and additional factors associated with an increased risk of venous thrombosis**Diseases and conditions**

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
 Behcet's disease
 Inflammatory bowel disease
 Nephrotic syndrome
 Limb paralysis
 Surgery

Additional risk factors for venous thromboembolism

Increasing age
 Hormone use
 Oral contraception
 Hormone replacement therapy
 Pregnancy and the puerperium
 Dehydration
 Immobility
 Due to illness
 Due to splinting
 During travel
 Medications
 Chemotherapy
 Tamoxifen
 Heparin (in heparin-induced thrombocytopenia)
 Indwelling venous catheters and devices

INTRODUCTION

Advances in understanding the pathogenesis of thrombosis have led to a massive increase in laboratory investigations to identify causal factors in affected individuals. However, this approach to investigation, when employed indiscriminately, does not improve clinical management, may cause confusion, and is potentially wasteful of scarce resources. In this article I shall review the inherited and environmental factors contributing to thrombosis, and explore the clinical value of laboratory investigation for prothrombotic states in patients with thrombosis.

THE PATHOGENESIS OF THROMBOSIS

The pathogenesis of thrombosis is multifacto-

rial – there is no single ‘cause’. Furthermore, the genetic, environmental and disease factors that contribute to venous thrombosis differ substantially from those that underlie arterial thromboembolism. In neurological practice, the emphasis is on arterial thrombosis and its consequences. However, deep venous thrombosis of the legs and pulmonary embolism frequently complicate the clinical course of neurological disorders, and intracranial venous thrombosis deserves special consideration.

VENOUS THROMBOSIS

Deep venous thrombosis occurs as a result of the interaction of multiple factors, both inherited and environmental. Out of Virchow's triad of causes of thrombosis, stasis and blood hypercoagulability predominate in relation to thrombosis in the deep veins of the lower limbs. Fibrin-rich thrombus forms in areas of sluggish flow. Vessel injury contributes in a minority of cases, only. In visceral and intracranial venous thrombosis, hypercoagulability may play a leading role, rather than stasis.

ARTERIAL THROMBOSIS

Arterial thrombosis is also multifactorial but here vessel wall pathology is central to the pathogenesis. Most frequently, platelet-rich thrombus forms on a ruptured or ulcerated atheromatous plaque and vessel occlusion or clot embolization leads to tissue infarction. Alternatively, there is embolism of clot formed in the fibrillating left atrium, or on damaged heart valves or left ventricular endocardium. Blood hypercoagulability may also contribute to the pathogenesis of arterial thrombosis.

Acquired situations and conditions that increase the risk of venous thrombosis have long been recognized (Table 1). The diseases and lifestyle factors that increase the risk of atherothrombosis are also familiar (Table 2).

THROMBOPHILIA

Recently, the term thrombophilia has been used to describe the situation in someone who is at increased risk of venous thromboembolism through an inherited predisposition. This followed the identification of some highly prevalent genetic risk factors for *venous* thrombosis, which result in a life-long procoagulant state and which contribute substantially to the burden of risk of venous thrombosis (Table 3). In

Table 2 Common factors and diseases associated with increased risk of atherothrombosis

Increasing age
Hypertension
Male sex
Diabetes mellitus
Dyslipidaemia
Obesity
Smoking
Sedentary lifestyle
Positive family history

Table 3 Thrombophilic conditions with a heritable component

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

contrast, the more prevalent of these genetic factors, for example factor V Leiden, make a negligible contribution to *arterial* thrombosis. Furthermore, despite considerable research in the area, the search for other prothrombotic genetic susceptibility markers for arterial thrombosis that make a *major* contribution to the burden of risk has not been fruitful. Most, such as polymorphisms in platelet receptor genes, carry a small or no risk (Table 4).

THE INHERITED THROMBOPHILIAS

Prior to 1994 the recognized genetic disorders known to carry increased risk of venous thrombosis were deficiencies of the physiological anti-

coagulants antithrombin, protein C and protein S. In that year a common polymorphism in the gene for factor V (factor V Leiden) was reported. It is associated with relative insensitivity to the anticoagulant action of protein C – ‘protein C resistance’ – and hence a procoagulant state (Figs 1 and 2). In the laboratory it is detected by the coagulation assay for the protein C resistant phenotype, or alternatively and more specifically, by detection of the point mutation using molecular genetic methods. Heterozygotes for factor V Leiden represent around 5% of the UK population and up to 10% in some northern European countries, and they have a background risk of venous thrombosis that is about fivefold greater than noncarriers. The risk is higher still in homozygotes. Whilst deficiency of antithrombin, protein C or protein S is detected in fewer than 10% of cases of venous thrombosis, factor V Leiden is present in up to around 40%. The detection rate is highest when those with a positive family history and apparently ‘spontaneous’ (not associated with an obvious temporary major risk factor such as surgery) thrombosis are tested. Although based on a limited number of cases, it seems that factor V Leiden also contributes to the occurrence of intracranial venous thrombosis.

More recently, another prothrombotic point mutation, in the gene for the coagulation factor prothrombin, has been reported. Prothrombin G20210A has a prevalence of around 1% in the UK. Up to 10% of patients with deep venous thrombosis are heterozygous for the gene. Predictably, it has been found in a similar proportion of individuals with intracranial venous thrombosis.

The heritability of thrombophilia also depends on the genes that determine the plasma concentrations of the clotting factors VIII, IX, XI and fibrinogen. The risk of venous thrombosis rises with their plasma concentrations. However, environmental factors also determine

Table 4 Candidate haemostatic markers and genetic risk factors for arterial thrombosis

Raised fibrinogen concentration
Raised factor VII concentration
Raised factor VIII concentration
Raised von Willebrand factor concentration
Platelet receptor gene polymorphisms
Thrombomodulin gene polymorphisms
Plasminogen activator inhibitor-1 (PAI-1) gene polymorphisms

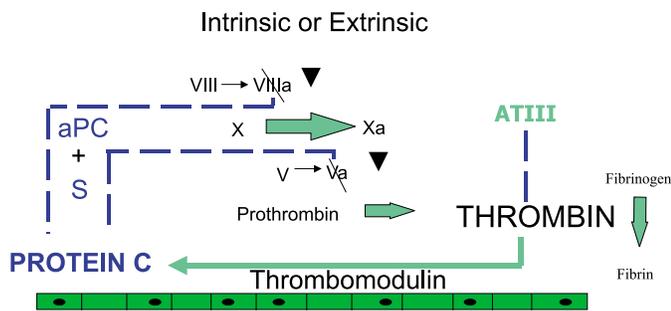


Figure 1 The principal physiological anticoagulant mechanisms. Thrombin, the enzyme responsible for conversion of soluble fibrinogen to insoluble fibrin, can also initiate an antithrombotic mechanism. When thrombin (generated through the coagulation cascade via the intrinsic or extrinsic pathway) binds to thrombomodulin on the surface of vascular endothelium. It activates protein C. Activated protein C, with its cofactor protein S, inactivates clotting factors V and VIII. This acts as a brake on further thrombin generation. Therefore deficiency of protein C or protein S, or of the inhibitor of thrombin itself (antithrombin III), results in a prothrombotic state. aPC, activated protein C; S, protein S; ATIII, antithrombin. Inhibitors in blue; blue broken line indicates inhibition.

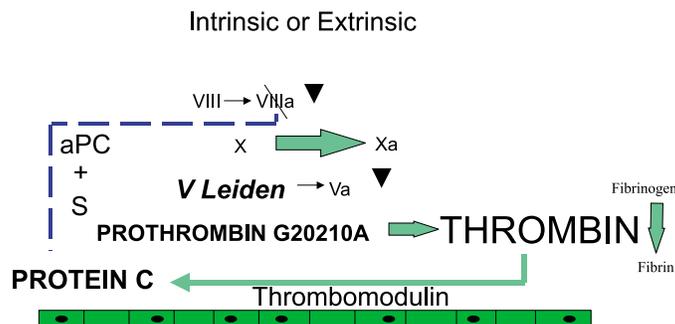


Figure 2 Factor V Leiden and prothrombin G20210A. The point mutation in the factor V gene results in synthesis of a variant of factor V that is insensitive to inhibition by activated protein C/protein S. Unimpeded activated factor V supports an increase in the background level of thrombin generation. The point mutation in the prothrombin gene results in enhanced synthesis and a higher plasma concentration of prothrombin, the zymogen for thrombin. aPC, activated protein C.

the plasma 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) as well as 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, which is associated with higher levels of homocysteine, is recognized, the mutation alone does not appear to convey increased risk of venous thrombosis. This suggests that other influences on homocysteine metabolism may be more important, such as folate status. There is no proven value in measuring homocysteine level in a patient with thrombosis at present because trials of the effect of suppression of plasma homocysteine on the risk of thrombotic events have not yet been completed.

Because factor V Leiden and prothrombin G20210A are quite common, coinheritance with other thrombophilias occurs. The risk of venous thrombosis is increased further in such cases, for example when factor V Leiden and protein C deficiency occur together in the same individual.

COAGULATION RISK FACTORS FOR ARTERIAL THROMBOSIS

There is little evidence to implicate factor V Leiden, prothrombin G20210A and deficiencies of protein C, protein S and antithrombin in *arterial* thrombosis. Exceptions may be rare situations, such as myocardial infarction in young women with other cardiovascular risk factors. A contribution to childhood stroke has been suggested, but is disputed. Although these genetic risk factors for venous thrombosis make little contribution to arterial disease, there are some associations between coronary and cerebrovascular thrombosis and coagulation factor levels. Fibrinogen is foremost among these. It is a likely candidate because it is the soluble substrate for thrombin in the generation of fibrin, it is an essential cofactor in platelet aggregation, and it makes a substantial contribution to the viscosity of blood. Plasma fibrinogen concentration

has been independently associated with the risk of stroke and myocardial infarction. Those with the highest levels have around a twofold increased risk. However, fibrinogen levels are highly-correlated with other cardiovascular risk factors, such as age and smoking, and so far there is no evidence that lowering fibrinogen concentration is protective and it is not easily achievable anyway. Furthermore, fibrinogen is an acute phase reactant and an increased plasma concentration could therefore merely reflect the presence of established atheromatous disease. Therefore, at present there is doubt about any cause and effect relationship between fibrinogen and arterial thrombosis. Similar considerations apply to a physiological inhibitor of fibrinolysis, plasminogen activator inhibitor (PAI-1). Although high levels are associated with atherothrombotic disease, PAI-1 is an acute phase reactant, its plasma level correlates with other risk factors, and cause and effect have not been demonstrated. Therefore, measurement of plasma fibrinogen and PAI-1 concentrations is of no value in patient management.

Other coagulation risk factors that have received attention as candidate markers for arterial thrombotic disease are coagulation factor VIII and von Willebrand factor (vWF), and factor VII. The best evidence is for factor VIII and vWF. The plasma concentrations are highly correlated, because they form a complex, and high levels in patients with atherosclerotic disease appear to predict increased risk of acute thrombotic events. However, again, knowledge of the plasma concentrations of factor VIII and vWF does not influence clinical management.

Although candidate genetic haemostatic risk factors for arterial thrombosis have been assessed (Table 4), in general the results are not conclusive. In many cases, for example some platelet receptor polymorphisms, there is no clear functional link between genotype and thrombosis. At present, there is no clinical benefit to the individual with heart attack or stroke from seeking these supposedly prothrombotic genes.

GENE-ENVIRONMENT INTERACTION

Patients with inherited thrombophilia, even those with combined defects, do not suffer from venous thrombosis most of the time. A trigger is usually necessary. Increasing age makes a significant contribution, through as yet unrecognized mechanisms. Other precipitat-

ing factors are listed in Table 1. The use of the combined oral contraceptive is worthy of special comment. In all users it induces a degree of resistance to the anticoagulant effect of protein C. This is comparable to the heterozygous state for factor V Leiden and results in an increased risk of venous thromboembolism of around four times background risk. However, as most users are young the absolute risk remains low. In women who are also heterozygous for factor V Leiden there is a multiplicative, rather than additive, interaction with the oral contraceptive, resulting in a 30- to 40-fold increased risk over background. Despite this, most such women still do not suffer thrombosis.

This, often multifactorial, gene-environment interaction, must underlie most episodes of venous thromboembolism. There is a combined effect of genetic predisposition (with some common thrombophilic variants, some less common and some not yet identified), interacting with increasing age and the influence of transient environmental or disease risk factors, which finally triggers an acute episode.

ANTIPHOSPHOLIPID SYNDROME

Systemic diseases contribute significantly to the occurrence of venous thrombosis. The antiphospholipid syndrome and cancer, including occult tumours, stand out because of their relatively high prevalence and an especially high risk of recurrent thrombosis. In the antiphospholipid syndrome, the presence of persistent antiphospholipid antibody is accompanied by thrombosis and/or recurrent pregnancy failure. There are other associations, including thrombocytopenia and livedo reticularis. In contrast to inherited thrombophilia, both arterial and venous thromboembolism are common. Ischaemic stroke, often occurring at a young age and in the absence of the usual risk factors, is an important feature. Venous thrombosis may affect unusual vessels, including intracranial veins. Laboratory diagnosis requires the demonstration of persistent anti-cardiolipin antibody or lupus anticoagulant. In some cases both types of antibody are present.

INTRACRANIAL VENOUS THROMBOSIS

In intracranial venous thrombosis, infection of neighbouring structures such as the middle ear has long been recognized as a cause. Unsurprisingly, additional provoking factors are similar to those in limb deep venous thrombosis. Use

of oral contraceptives, pregnancy, dehydration, polycythaemia, sickle cell disease, malignancy, paroxysmal nocturnal haemoglobinuria, the antiphospholipid syndrome and inflammatory bowel disease have all been implicated. Inherited thrombophilia is probably present in a similar proportion to that in patients with limb deep venous thrombosis.

THE DIAGNOSIS OF THROMBOPHILIA

It has become commonplace to seek laboratory confirmation of inherited thrombophilia in almost everyone with acute venous thromboembolism. Although laboratory tests for the inherited thrombophilias are readily available, their clinical value is far from clear. Furthermore, testing should not detract from the consideration of important, often modifiable, environmental and disease risk factors, which may be contributing to the pathogenesis of the thrombotic episode.

Thrombophilia testing at presentation of acute venous thrombosis of the limbs

Identification of inherited thrombophilia is seldom, if ever, of value in the management of the acute episode. This is because the genetic predisposition has no influence on the severity nor extent of thrombosis, nor on the response to anticoagulant therapy. Furthermore, even the duration of anticoagulant therapy is not usually influenced by the results of tests for inherited thrombophilia. This is because the recurrence rates of limb deep venous thrombosis on discontinuing anticoagulant therapy are no different in factor IV Leiden heterozygotes from those in whom inherited thrombophilia is not detected. Indeed, there is no reason why they should be. The occurrence of venous thromboembolism in an individual in circumstances when most do not suffer from an event identifies that person as predisposed, whatever the results of the laboratory tests. An exception may be antithrombin deficiency, where clinical anecdote suggests higher recurrence rates. This may also be the case for homozygotes for factor V Leiden and prothrombin G20210A, double heterozygotes, and those with combined thrombophilic defects. However, these individuals are encountered only rarely and tend to present with thrombosis at an early age, often with a strong family history. For these reasons it is appropriate to be selective in the investigation for inherited thrombophilia. Priority should be

given to the young, those with two or more first degree relatives with venous thrombosis, and patients in whom there is no clear environmental factor or disease that has provoked the acute episode. Even then there is no evidence to guide therapeutic decisions based on the detection or otherwise of inherited thrombophilia.

Intracranial venous thrombosis and ischaemic arterial stroke

There is no evidence that any different considerations to venous thrombosis in the legs should apply to intracranial venous thrombosis. Furthermore, because the inherited thrombophilias play little if any part in arterial thrombosis, there is no value in routine testing for these conditions in ischaemic arterial stroke. Indeed, such an approach may lead to unjustifiable interventions because of the high prevalence of thrombophilic genotypes in the general population: heterozygosity for factor V Leiden or prothrombin G20210A will be detected by chance in around 6% of cases of ischaemic arterial stroke.

The diagnosis of inherited thrombophilia and the prevention of venous thromboembolism

The likely impact of the increased knowledge of inherited thrombophilias is in the prevention of venous thromboembolism in clinically-unaffected carriers. However, this advance has yet to be realized. Screening of populations at risk has been advocated but the effectiveness of such a policy is open to doubt. For example, although there is a multiplicative effect between factor V Leiden and oral contraceptive use, the background rate of venous thrombosis in the young and fit population in question is very low. Thus it has been calculated that several million women must be screened to prevent a single fatal event. Furthermore, some women would avoid oral contraception as a result of a positive test with the inevitable increased risk of pregnancy. The situation is similar in screening women prior to hormone replacement use, although the background rate of thrombosis is higher in this older population.

Case finding to permit family studies in order to identify relatives at increased risk is likely to be a more effective approach to thrombosis prevention. However, most heterozygotes for factor V Leiden will never suffer venous thrombosis and the psychological and lifestyle risks associated with the diagnosis of a genetic predisposi-

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tion should not be underestimated. Testing for inherited thrombophilia should not be used indiscriminately, and appropriate facilities for counselling before and after testing must be in place. Even then, benefit is likely mainly from testing in families where there is a history of venous thrombosis in more than one individual, especially when events occur at a relatively young age, and are apparently unprovoked.

The problem of diagnostic error – pitfalls in the laboratory diagnosis of inherited thrombophilia

A further complication stems from the possibility of diagnostic error. This is especially relevant to deficiencies of proteins C and S, because with the commonly-employed assays there is overlap between levels in heterozygotes and unaffected individuals. Also the plasma concentration of anti-thrombin falls during acute thrombotic episodes and with heparin therapy. Furthermore protein C and protein S are vitamin-K-dependent proteins and deficiency cannot be diagnosed reliably during warfarin therapy. Their levels also change during normal pregnancy. Finally, if the protein C resistance assay is used to test for factor V Leiden, confusion may arise because protein C resistance may be a consequence of atherothrombosis. Great care is required therefore in the choice and interpretation of tests and in the timing of blood sampling for testing for thrombophilia.

Particular issues arise in the performance of genetic tests for a generally late onset disorder with a low fatality rate and incomplete penetrance. In investigating for thrombophilia it is essential to consider what can be achieved and whether harm can result. Harm includes generation of anxiety in asymptomatic relatives, unwanted pregnancy due to avoidance of oral contraception, unnecessary denial of the benefits of hormone replacement therapy and inappropriate exposure to antithrombotic therapies.

FURTHER READING

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PUTTING CURRENT KNOWLEDGE INTO CLINICAL PRACTICE

Based on the considerations presented in this article the conclusions are:

- Thrombosis is a multihit condition – there is no single ‘cause’.
- In venous thromboembolism in the legs and in the head the clinical history and physical examination will often reveal important modifiable risk factors.
- Inherited thrombophilia is not modifiable and detecting it should rarely influence clinical management; therefore its detection can be regarded as of secondary importance.
- If testing for inherited thrombophilia is pursued it is only likely to be of benefit if facilities are in place for testing at risk relatives as well. The provision of expert counselling is essential. Usually this will require liaison with haematologists and clinical geneticists. Whether this is a cost effective approach to thrombosis prevention has not been established.
- There is no indication for thrombophilia testing during the acute presentation with thrombosis because the results may be misleading.
- In arterial thrombosis, including ischaemic stroke, the clinical history and physical examination, along with simple biochemical tests will often reveal important modifiable risk factors
- In ischaemic stroke, routine testing for inherited thrombophilia is not indicated because the common thrombophilic conditions do not contribute significantly to the pathogenesis, and treatment would not be affected.
- There is no benefit in seeking evidence of alterations in coagulation factors and genetic susceptibility genes in an individual with stroke because management would not be affected.
- In patients with arterial or venous thrombosis, antiphospholipid antibodies should be sought as the thrombosis recurrence rate in the antiphospholipid syndrome is particularly high. Longer term antithrombotic therapy may be indicated, although the substantial bleeding risk from warfarin should be carefully considered.