<table>
<thead>
<tr>
<th>Title of Guideline (must include the word “Guideline” (not protocol, policy, procedure etc))</th>
<th>Guideline for requesting thrombophilia testing</th>
</tr>
</thead>
</table>
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Associate Specialist in Haematology |
| Directorate & Speciality | Pathology |
| Date of submission | July 2015 |
| Date on which guideline must be reviewed (this should be one to three years) | July 2023 |
| Explicit definition of patient group to which it applies (e.g. inclusion and exclusion criteria, diagnosis) | Individuals with a personal or family history of venous thrombosis. |
| Abstract | This guideline provides evidence based recommendations for thrombophilia testing to try and ensure that it is targeted at the right patients. |
| Key Words | Thrombophilia; venous thrombosis; |
| Statement of the evidence base of the guideline – has the guideline been peer reviewed by colleagues? | Based on: |
| Evidence base: (1-5) | NICE clinical guideline June 2012: Venous thromboembolic diseases: the management of venous thromboembolic diseases and the role of thrombophilia testing. |
| | Clinical guidelines for testing for heritable thrombophilia, British Committee for Standards in Haematology (December 2009) |
| | Reducing the Risk of Venous Thromboembolism during Pregnancy and the Puerperium RCOG Green-top Guideline No. 37a 2015 |
| | Peer reviewed by multi-disciplinary team of specialists in haemostasis and thrombosis comprising clinicians, specialist nurses, clinical and biomedical scientists. |
| Consultation Process | Consultation with a cross section of clinicians in primary and secondary care currently requesting thrombophilia testing identified from laboratory audit. |
| Target audience | Clinicians within the Trust and GPs. |

This guideline has been registered with the trust. However, clinical guidelines are guidelines only. The interpretation and application of clinical guidelines will remain the responsibility of the individual clinician. If in doubt contact a senior colleague or expert. Caution is advised when using guidelines after the review date.
Guideline for requesting for thrombophilia testing

1. INTRODUCTION

1.1. Purpose of the guideline

It is important to both patients and clinicians that thrombophilia testing is appropriately targeted. This guideline outlines local recommendations for thrombophilia testing at this Trust.

The term "thrombophilia" means an inherited or acquired predisposition to venous thromboembolism (VTE). There are a number of laboratory tests for thrombophilic abnormalities, although it is recognised that currently available ‘thrombophilia screens’ cannot fully assess an individual’s thrombotic potential. Individual risk is affected by multiple genetic and environmental factors which will be different even amongst first degree relatives.

Whilst it is natural for both patient and clinician to want to look for the cause of a thrombosis, it is recommended only to perform thrombophilia testing where the management of the patient will be altered by the results.

It is important to realise the limitations of thrombophilia testing.

- The patient’s personal and family history of thrombosis are almost always more important than the thrombophilia test result and a negative result should not be allowed to provide false reassurance for the patient and clinician.
- Unless testing for a specific heritable thrombophilic defect in a family known to carry that defect, a negative result does not exclude the possibility of an inherited predisposition to thrombosis. There are known to be families with a clear predisposition to thrombosis in whom a thrombophilic abnormality has not been demonstrated by currently available tests. It is likely that these families carry an as yet undetected familial thrombophilia, which may be discovered in the future. Thrombophilia testing in such families will not be helpful and a negative result may offer false reassurance for patients and clinicians.
- Testing for heritable thrombophilia does not in many cases predict likelihood of recurrence in VTE patients and does not reduce recurrence rates.
- Decisions regarding duration of anticoagulation (lifelong or not) in unselected patients should be made with reference to whether or not a first episode of venous thrombosis was provoked or not, other risk factors, and risk of anticoagulant therapy-related bleeding, regardless of whether a heritable thrombophilia is known.
- In many families there is a low risk of thrombosis in asymptomatic relatives. The results of thrombophilia tests are frequently misinterpreted and may lead to anxiety and distress for otherwise healthy individuals.

1.2. Target audience

Clinicians within the Trust and GPs.
2WHEN, AND IN WHOM SHOULD THROMBOPHILIA TESTING BE DONE?

Thrombophilia testing is expensive and time-consuming. Unfortunately, testing is often performed without a clear idea of what to do with the results.

Positive results often cause unjustified concern to the individual. Conversely, a negative test may be used to provide false reassurance.

Therefore it is important that testing is targeted at the right people and the requesting clinician must have a plan of how the results will affect management.

There is no evidence to support testing for thrombophilia in patients who are continuing anticoagulant therapy indefinitely after an episode of VTE. This is because the results of such testing do not influence patient management.

In individuals with unprovoked VTE, testing for anti-phospholipid syndrome must be considered, especially if cessation of anticoagulation is being considered. This is because confirmation of the presence of APS will definitely influence management. Although anti-cardiolipin and beta 2 glycoprotein 1 antibodies can be tested at any time, testing for lupus anticoagulant may not be reliable in patients taking warfarin.

A summary of the recommendations can be found in Appendix 1.

2.1 Request screening

All requests for thrombophilia testing are screened by a senior haematologist.

If clinical details are absent or incomplete, or the request is deemed inappropriate, samples will be saved by the laboratory for 2 weeks, to enable the clinician to provide further information and/or discuss the case with a haematologist.

In the event that a request for thrombophilia testing is deemed inappropriate, a comment will be given on the report, and a web link to this guideline will be provided.

It is recommended that clinicians discuss difficult cases with a senior haematologist, or refer to the Thrombophilia clinic for further advice.

2.2 Limited testing

In some cases limited testing eg screening for anti-phospholipid syndrome will be appropriate when other thrombophilia tests are not. Again, a relevant comment will be made with the results and the sample will be saved pending further communication with the clinician.

2.3 Timing of the test

- Full thrombophilia testing should not be carried out during the acute presentation with thrombosis as the thrombotic process itself may affect the results and testing at this time does not influence management
• Testing for anti-phospholipid syndrome may be appropriate in some clinical circumstances during an acute presentation.
• Full thrombophilia testing should be delayed until 4-6 weeks after stopping anticoagulant therapy.

2.4 Specific indications for thrombophilia testing

**Full testing** (see 4 below for current available tests) will be performed without further haematological review in the following circumstances providing relevant clinical details are given on the request form.

• Venous thrombosis at unusual site eg cerebral, abdominal. (Testing for acquired conditions such as paroxysmal nocturnal haemoglobinuria (PNH) and testing for the JAK 2 mutation associated with myeloproliferative neoplasms is also advised.)

It is strongly recommended that patients in whom testing for heritable thrombophilia is being considered are referred to the thrombosis clinic, or discussed with a haematologist.

**Limited thrombophilia testing** will be appropriate in the following cases:

• 1\textsuperscript{st} episode of unprovoked VTE in patients for whom it is planned to stop anticoagulant therapy
  - Lupus anticoagulant, (and anticardiolipin antibodies, anti-\(\beta_2\) glycoprotein 1 antibodies if clotted sample provided)

• Warfarin induced skin necrosis
  - Protein C and S only

• Neonatal purpura fulminans
  - Protein C and S only

• Arterial thrombosis
  - Lupus anticoagulant (and anticardiolipin antibodies, \(\beta_2\)GP1 if clotted sample provided)

• Patient with NO personal history of thrombosis but with family history tracking with a **specific high risk thrombophilic abnormality**
  - **Test only for known high risk defect** (Antithrombin, Protein C or Protein S). It is recommended that these individuals are referred for assessment in the thrombophilia clinic.

2.5 Situations in which thrombophilia testing MAY be considered

Decisions regarding duration of anticoagulation and assessment of risk of recurrent VTE should be made based on an assessment of possible provoking factors for the index event; ongoing risk factors for thrombosis and the severity of the original event. Requests for testing for heritable thrombophilia in the following patient groups will not automatically be processed.

**We strongly recommend that these patients are referred for assessment by a haematologist prior to the request for testing to avoid delays, anxiety and unnecessary venepuncture.**

A comment to this effect and a link to these guidelines will be added to the report.

• First episode of unprovoked venous thrombosis in patients who have a first-degree relative who has had DVT or PE, if it is planned to stop anticoagulation treatment.
• First episode of unprovoked VTE in patients < 50 years who have NO family history of VTE, and in whom anticoagulation would otherwise be discontinued.
For many patients in the above categories, thrombophilia testing is not of any clinical utility as it does not influence their management (particularly testing for the lower risk abnormalities).

- Patients < 50 years with provoked VTE who have a first degree relative with VTE.
- Patients with provoked VTE at a very young age.

*It is unlikely that thrombophilia testing will be recommended for most patients who have had provoked VTE as these patients are at less risk of recurrence and will be given short-term anticoagulation as standard treatment whether they have thrombophilia or not. Testing therefore has no utility as it does not change patient management.*

2.6 Children

Thrombophilia testing is very rarely indicated in childhood. In addition, robust reference ranges for children are not available and results can be very difficult to interpret. All requests for thrombophilia testing in a child should first be discussed with a paediatric haematologist. Requests will only be processed if the clinician indicates on the request form which paediatric haematologist has recommended testing.

2.7 Pregnancy morbidity/ thromboprophylaxis in pregnancy

The Royal college of Obstetrics and Gynaecology recently published revised guidelines regarding thromboprophylaxis in pregnancy. This included specific recommendations on thrombophilia testing. In addition the British Committee for standards in Haematology have also published guidance in this setting.

- Women should be assessed for risk of pregnancy-associated venous thrombosis primarily in relation to clinical risk factors.
- Prior to testing for thrombophilia, women should be counselled regarding the implications for themselves and family members of a positive or negative result. The results should be interpreted by clinicians with specific expertise in the area.
- Most pregnant women with a previous unprovoked venous thrombosis or hormone related thrombosis will qualify for thromboprophylaxis during pregnancy based on clinical risk alone and so testing for heritable thrombophilia is usually not required except: Pregnant women with prior VTE who also have a family history of antithrombin deficiency or where the specific thrombophilia has not been detected should be tested for antithrombin deficiency alone
- Pregnant women with a previous event due to a major provoking factor, e.g. surgery or major trauma do not require testing
- Women with an unprovoked VTE should be tested for the presence of antiphospholipid antibodies.
- Women with no personal history or risk factors for VTE but who have a family history of an unprovoked or oestrogen-provoked VTE in a first-degree relative when aged under 50 years should be considered for thrombophilia testing. This will be more informative if the relative has a known thrombophilia.
- Antithrombotic therapy should not be given to pregnant women with a history of pregnancy complications/poor obstetric history based on testing for heritable thrombophilia; therapeutic decisions should be based on clinical circumstances.
- Protein S levels decrease in pregnancy, and therefore should not be tested.
2.8 Other requests not covered above

To avoid unnecessary venepuncture and delays in processing requests, it is recommended that any other requests not covered by this guideline should be discussed in advance with a haematologist, or referred to the Thrombophilia clinic.

3 WHEN IS TESTING FOR HERITABLE THROMBOPHILIA UNLIKELY TO BE HELPFUL?

3.1 Combined Oral Contraceptive Pill / Hormone Replacement Therapy

Any woman wishing to take the Combined Oral Contraceptive Pill (COCP) or Hormone Replacement Therapy (HRT) who has a family history of VTE in a first degree relative should consider an alternative non-oestrogen containing form of contraception, or transdermal HRT.

If the affected relative has not had thrombophilia testing, or does not have a measurable thrombophilic abnormality, testing in the asymptomatic patient will provide an uncertain estimate of risk and is not recommended.

If a symptomatic first degree relative has been tested and has a measurable heritable thrombophilia, testing asymptomatic relatives may not necessarily be helpful, as a negative test does not exclude an increased risk of VTE. Non-oestrogen alternatives should still be considered first.

Testing for heritable thrombophilia may assist counselling of some selected women, particularly if a high risk thrombophilia has been identified in a symptomatic relative. In these cases it is recommended that the individual is referred to the Thrombophilia clinic for counselling and possible testing.

3.2 Arterial thrombosis

No heritable components of the current ‘Thrombophilia Screen’ have been firmly established as risk factors for arterial thrombotic diseases.

Treatment and secondary prevention should be in relation to established cardiovascular risk factors. Testing for heritable thrombophilia is NOT recommended, and will only be performed after discussion with a senior haematologist.

However anti-phospholipid syndrome may present with arterial thrombosis and testing for the lupus anticoagulant, anticardiolipin and β2GP1 antibodies may be indicated in patients with arterial thrombosis.

3.3 Screening asymptomatic relatives of individuals with low risk heritable thrombophilia

Case finding of asymptomatic relatives with low risk thrombophilia, such as the Factor V Leiden and Prothrombin Gene mutations, is not recommended as it has not been shown to reduce the incidence of venous thrombosis and the annual risk of unprovoked thrombosis in affected family members is low. Testing in these circumstances may cause unnecessary anxiety to patients.
Testing may be appropriate in some families affected by high risk heritable thrombophilia (Antithrombin, Protein C or Protein S deficiency). It is recommended that such individuals are referred for assessment in the Thrombophilia clinic.

4 CURRENTLY AVAILABLE THROMBOPHILIA TESTS

Components of a full thrombophilia ‘screen’:

**a) Tests for inherited thrombophilia**

- **Antithrombin** is a major inhibitor of blood coagulation and is essential for effective heparin therapy. It inhibits the coagulation proteases including IIa, IXa, Xa and XIa. **Antithrombin deficiency** is very rare (prevalence 0.02%) but has a high risk of venous thrombosis.

- **Protein C** is a vitamin K-dependent natural anticoagulant protein made by the liver. It is converted to activated protein C (APC) by thrombin. APC inactivates factors Va and Vlla. **Protein C deficiency** is rare (prevalence 0.2%) but has a moderate - high risk of venous thrombosis.

- **Protein S** is a vitamin K-dependent protein made by the liver. It is the cofactor for the anticoagulant activity of APC. It circulates in a free form (40%) or bound to the acute phase C4b-binding protein (60%). Only the free form is functional and only this is measured in the thrombophilia screen. **Protein S deficiency** is rare (prevalence 0.03-0.13%) but has a moderate - high risk of venous thrombosis.

  *For all three natural anticoagulants, environmental factors may lead to acquired deficiency.*

  - Severe liver disease reduces antithrombin, protein C and protein S.
  - Severe vitamin K deficiency, usually due to warfarin therapy, reduces protein C and protein S.
  - Pregnancy and oestrogen therapy both reduce protein S. Protein S falls very early in pregnancy and low protein S can persist for 6-8 weeks even after early miscarriage.

- **Factor V Leiden variant** \((F5\,c.1601G>A\, (p.Arg534Gln))\) produces a factor V molecule that is resistant to cleavage by Activated Protein C. FVL is tested by MS-PCR, together with the prothrombin variant. FVL is the most prevalent thrombotic risk factor known in the Caucasian population (around 5%). Heterozygotes have a modest increase in the risk of thrombosis. Homozygotes are much less common but have a much higher thrombotic risk.

- **Prothrombin Gene variant** \((F2\,g.20210G>A)\) causes elevated levels of prothrombin in the circulation. This variant is tested by MS-PCR together with FVL. Heterozygotes are common in the Caucasian population (around 3%) and have a small increased risk of thrombosis. Homozygotes or compound heterozygotes with FVL have a significantly greater risk of thrombosis.

**b) Tests for acquired thrombophilia**

- **Lupus Anticoagulant,** anti-cardiolipin antibodies and anti β2GP1 antibodies are present in the Anti-Phospholipid Syndrome (APS), an **acquired** thrombophilic state. APS is an autoimmune disease characterised by arterial and/or venous thrombosis or pregnancy complications in the presence of persistent anti-phospholipid antibodies.
Lupus anticoagulant is a clotting based test which can be performed on the same sample as the tests for heritable thrombophilia. A separate sample must be sent to Immunology to test for anti-cardiolipin and anti β2GP1 antibodies.

5. REFERENCES

1. NICE clinical guideline June 2012: Venous thromboembolic diseases: the management of venous thromboembolic diseases and the role of thrombophilia testing.

2. Thrombophilia, British Heart Foundation (Factfile), 2002.


4. Clinical guidelines for testing for heritable thrombophilia, British Committee for Standards in Haematology (December 2009)


8. Reducing the Risk of Venous Thromboembolism during Pregnancy and the Puerperium RCOG Green-top Guideline No. 37a April 2015
**Appendix 1: Summary of recommendations**

<table>
<thead>
<tr>
<th>Thrombophilia testing RECOMMENDED:</th>
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<tbody>
<tr>
<td>1. Neonatal purpura fulminans</td>
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<tr>
<td>2. Warfarin induced skin necrosis (Protein C and S only)</td>
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<tr>
<td>3. VTE at unusual site eg cerebral, abdominal</td>
</tr>
<tr>
<td>4. Testing for anti-phospholipid syndrome ONLY should be strongly considered in all patients with first episode of unprovoked VTE if it is planned to stop anticoagulant therapy.</td>
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<tr>
<td>5. Arterial thrombosis where antiphospholipid syndrome is a possibility (Lupus anticoagulant, anticardiolipin antibodies and β2GP1 antibodies only)</td>
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<table>
<thead>
<tr>
<th>Thrombophilia testing MAY BE CONSIDERED Refer to thrombosis clinic to consider testing:</th>
</tr>
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<tbody>
<tr>
<td>1. First episode of unprovoked VTE in patients who have a first-degree relative who has had DVT or PE if it is planned to stop anticoagulation treatment</td>
</tr>
<tr>
<td>2. First episode of unprovoked VTE in patients &lt; 50 years who have NO family history of VTE, and in whom anticoagulation would otherwise be discontinued.</td>
</tr>
<tr>
<td>3. Patients &lt; 50 years with provoked VTE who have a first degree relative with VTE.</td>
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<tr>
<td>4. Patients with provoked VTE at a very young age.</td>
</tr>
<tr>
<td>5. Asymptomatic relatives of patients with known high risk thrombophilia (AT, Protein C, Protein S). Appropriate counselling essential.</td>
</tr>
<tr>
<td>6. Asymptomatic relatives of VTE patients with known high risk thrombophilia considering oestrogen containing contraception/HRT (estimate of risk uncertain. Appropriate counselling essential)</td>
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<table>
<thead>
<tr>
<th>Thrombophilia testing NOT RECOMMENDED:</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Acute VTE</td>
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<tr>
<td>2. Patient receiving anticoagulants (discuss with a haematologist)</td>
</tr>
<tr>
<td>3. Arterial thrombosis, except where APS is suspected (see above)</td>
</tr>
<tr>
<td>4. Asymptomatic relatives of patients with low risk thrombophilia</td>
</tr>
<tr>
<td>5. Decisions about oestrogen containing contraception/HRT (for exception, see above)</td>
</tr>
<tr>
<td>6. Children (discuss with paediatric haematologist)</td>
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<tr>
<td>7. Retinal vein thrombosis</td>
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<tr>
<td>8. Central venous catheter related thrombosis</td>
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# Appendix 2

## Heritable risk factors for thrombosis – incidence and relative risk

<table>
<thead>
<tr>
<th>Thrombophilia</th>
<th>Prevalance in Caucasian population (%)</th>
<th>Relative risk of venous thromboembolism (VTE)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Factor V Leiden (FVR506Q or activated protein C resistance)</td>
<td>5.0 (varies from 0-15% in different populations)</td>
<td>heterozygotes 3-8 x homozygotes 80 x</td>
</tr>
<tr>
<td>Prothrombin G20210A</td>
<td>2.0</td>
<td>3 x</td>
</tr>
<tr>
<td>Protein C deficiency</td>
<td>0.2 (estimated)</td>
<td>10-15 x</td>
</tr>
<tr>
<td>Protein S deficiency</td>
<td>0.03-0.13 (estimated)</td>
<td>10 x</td>
</tr>
<tr>
<td>Antithrombin deficiency</td>
<td>0.02</td>
<td>25-50 x</td>
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</table>