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Acknowledgements

I would like to thank the following people for their counselling, encouragement and efforts during the preparation of this commentary:

Professor John Anthony FCOG(SA), Head of Maternity Services, Department of Obstetrics and Gynaecology, Groote Schuur Hospital for supervising the project.

Dr Rosie Burton MRCOG, PhD. Ex. Perinatal Fellow, Department of Obstetrics and Gynaecology, Groote Schuur Hospital for her assistance with the research proposal and setting up the research study.

Professor Nicolas Novitzsky, Head of Department of Haematology and the rest of the haematology laboratory team for their assistance in running the haematology investigations.

Mrs Megan Howes for her assistance with lay-out and formatting.
This is to certify that this commentary is the sole and exclusive work of the candidate.

Signed by candidate

Dated at London, UK

Dr Vincent Boama
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INTRODUCTION

Venous thromboembolism (VTE) is a major cause of maternal mortality worldwide. In many developed countries, all maternal deaths are investigated, and accurate statistics are available. In United Kingdom (UK) for example, VTE is the leading cause- and is increasing despite heightened awareness of risk factors, and wider use of thromboprophylaxis\(^{(1,2)}\). The 1994-96 UK Confidential Enquiries reported an overall maternal mortality rate of 12.2 per 100,000 deliveries, with specific mortality from VTE at 2.2 per 100,000 deliveries, with approximately 15 deaths a year due to VTE\(^{(1)}\). Similarly the 1997-1999 UK confidential enquiries reported an overall maternal mortality of 11.4 deaths per 100,000 maternities with thrombosis and thromboembolism remaining the major cause of death at a rate of 16.5 per million maternities \(^{(2)}\).

In developing countries, maternal mortality is substantially higher, and there are considerable difficulties in collecting verifiable statistics. The total number of deliveries per annum may be unknown. Maternal deaths may be underreported, and there may frequently be very little clinical or autopsy information available to establish the cause of death. Despite these difficulties, South Africa is now committed to producing an annual audit of maternal mortality\(^{(3)}\). The recently published 1998 Confidential Enquiries gave an overall estimated maternal mortality rate of 150 per 100,000 deliveries. Deaths from VTE contribute less to the overall total, being considerably outnumbered by death from hypertensive disorders and haemorrhage. There were 41 deaths attributed to "acute collapse and embolism" - in many of these cases little information was available concerning the etiology of acute collapse. Seven women died from proven pulmonary embolism. It is likely that many other women who died from
acute collapse as well as others categorised as "home deaths" of unknown etiology also resulted from VTE. Overall mortality from "acute collapse and embolism" was 4.9 per 100,000 estimated deliveries. If even half were due to pulmonary embolism, this would give a maternal mortality equivalent to that in the UK. Venous thromboembolism therefore appears to be a significant medical problem in pregnancy for women in South Africa.

**RISK FACTORS FOR VTE**

Venous thromboembolism is a multi-causal disorder, involving the interaction of genetic and acquired factors\(^4\). Pregnancy is a significant risk factor, increasing the risk of VTE by 5 fold. Other acquired risk factors for VTE are well known. These include prolonged immobility, increasing body mass index, major surgery, and use of the combined oral contraceptive pill\(^5\). Increasing age is also a significant risk factor.

Thrombophilia is defined as the predisposing tendency to the occurrence of thrombosis. In recent years, it has become apparent that a significant proportion of individuals with VTE have underlying inherited thrombophilia. These disorders result in either deficiencies of anticoagulant activity, or an increased procoagulant tendency. Several underlying genetic abnormalities are well characterised, and more are likely to follow. Many people with genetic thrombophilias are asymptomatic. In those who develop VTE, more than one inherited disorder is often present. Acquired risk factors such as pregnancy may act as a trigger for thrombosis in individuals with an increased genetic risk.

The prevalence of inherited thrombophilias varies considerably between different
populations. The majority of data in the published literature is from North America and Western Europe. There is very little information relating to other ethnic groups. Only one large study to date has investigated the prevalence of thrombophilias in South Africa. This included 350 patients with VTE, and found that the peak incidence occurred approximately 10 years younger than in North American and Western European populations\(^6\). This suggests that inherited thrombophilias may be important risk factors in the South African population. On the other there have been other few studies in South Africa that have investigated relevant polymorphism in the context of pre-eclampsia and eclampsia\(^7,8,9\). None of these studies did show any significant contribution from genetic thrombophilia to the pathophysiology of placental vasculopathy in black South Africans.

Thrombophilia may also be acquired. Hyperhomocysteinaemia may have a genetic basis, or be acquired due to vitamin deficiency. The autoimmune disorders Antiphospholipid Antibody Syndrome and Systemic Lupus Erythematosus are both associated with arterial and venous thrombosis.

INHERITED THROMBOPHILIA

(i) Decreased anticoagulant activity.

Naturally occurring anticoagulants limit the activity of the clotting cascade. Genetic polymorphisms leading to deficiencies of antithrombin, protein C and protein S have been described\(^10\). These are autosomal dominant, with heterozygotes having an increased risk of venous thromboembolism. Homozygotes are rare. Homozygous antithrombin deficiency leads to death in
infancy. Homozygous protein C and protein S deficiency results in severe, lifelong thrombotic disease. Multiple different mutations have been described for each deficiency; more than 80 are known resulting in antithrombin deficiency. Different mutations have different thrombotic tendencies; those that are the most thrombogenic will be detected most often in symptomatic patients. The risk of VTE in asymptomatic patients with thrombophilia is therefore difficult to ascertain.

Antithrombin is an inhibitor of the activated forms of factor II, IX, X, XI and XII. Additionally it is essential for heparin to exert its anticoagulant effect. Antithrombin is activated by heparin binding; it is therefore ineffective in individuals with antithrombin deficiency unless antithrombin concentrates are co-administered. Functional levels of greater than 50-70% are required to avoid an increased risk of thrombosis\(^{(10)}\). In European populations antithrombin deficiency has a reported prevalence of 0.1% in the general population. Overall, it is considered the most thrombogenic inherited thrombophilia, with at least a 50% lifetime risk of thrombosis. The prevalence of antithrombin deficiency in South Africa is unknown.

Protein C and protein S form part of the same anticoagulant system. Protein C is activated through binding to thrombin, which then inactivates factor Va and VIIIa via proteolytic cleavage. Protein S acts as a cofactor for this pathway. Mutations in factor V can lead to missing cleavage sites for protein C, resulting in a decreased sensitivity to inactivation. This phenomenon is termed activated protein C resistance (APCR). The most common molecular basis for this involves substitution of glutamine for arginine at position 506, known as the factor V Leiden mutation\(^{(11)}\). This disorder is also autosomal dominant, with homozygotes
having a greatly enhanced risk of thrombosis. To date mutations in factor VIII leading to APCR have not been described, but may be described in the future.

Inherited defects in the protein C system are more common than antithrombin deficiency in populations of European origin. The prevalence of protein C deficiency and protein S deficiency are 0.2% and 0.2-2% respectively. Factor V Leiden is the most common inherited thrombophilia in European populations, with a heterozygote frequency of around 5%. The prevalence of protein C and protein S deficiency is unknown in South Africa, or amongst Black African populations elsewhere. Factor V Leiden has been extensively studied in different populations. It is extremely rare in people of African and Asian descent\(^{(12)}\). A recent study in South Africa showed this mutation to be virtually absent amongst the Black population\(^{(6)}\).

**Increased Procoagulant Activity**

Less attention has been given to the association between increased concentrations of procoagulant factors and venous thromboembolism\(^{(13)}\). It is becoming increasingly apparent that these may also underlie an inherited tendency to thrombosis.

A genetic mutation in the gene for prothrombin (factor II) has been described that results in an increased risk of VTE\(^{(14)}\). This results in a guanine to arginine transition at position 20210 in the 3' untranslated region. Rather than resulting in a deficiency of the final protein product, gene regulation is disrupted, leading to increased levels of prothrombin. The frequency of this mutation is high in European populations. A Dutch study found a prevalence of 18% amongst
individuals from symptomatic families, compared to 1% amongst healthy controls. It is less thrombogenic than antithrombin deficiency, with an odds ratio for VTE of 2.8, (95% CI 6-30) \(^{(4)}\).

Recent research has established that factor VIII is an independent risk factor for thrombosis. Factor VIII concentrations > 150 IU/dl are associated with a 5-6 fold increased risk of venous thromboembolism compared to levels < 100IU/dl\(^{(15)}\). A further study found levels > 175 IU/dl (the 90\(^{th}\) centile for healthy controls) in 19% of people with a single episodes of VTE, and 33% of those with recurrent episodes\(^{(16)}\). A dose dependant effect was also revealed. For each further rise of 10IU/dl, the risk for single and recurrent episodes increased by 10% and 24% respectively. A molecular basis for increased factor VIII levels has yet to be described. However familial clustering has been documented, and other evidence suggests that factor VIII levels are genetically determined.

Elevated fibrinogen levels are also associated with an increased risk of VTE\(^{(15)}\). Compared to a baseline concentration of < 3.0g/l, fibrinogen >5.0 g/l increases the risk 3-4 fold. Concentrations above this threshold were found in 3% of patients with a first episode of VTE in a European study, compared to 1% of controls.

It is likely that raised concentrations of other procoagulant factors will be linked to an increased risk of VTE in the future. At present, both factor XI and factor XIII are under investigation. Factor XI levels above the 90\(^{th}\) centile have been associated with a 2.2 fold risk of VTE\(^{(17)}\). A common polymorphism of factor XIII appears to be associated with protection against VTE, with the wild type genotype found more frequently in individuals with a history of thrombosis\(^{(18)}\).
ACQUIRED THROMBOPHILIA

Antiphospholipid antibodies are strongly associated with thrombosis with an increased risk of both arterial and venous thrombosis. This is the most common form of acquired thrombophilia. In the United States of America, 25% of patients with venous thrombosis are antiphospholipid antibody positive. Anticardiolipin antibodies and the lupus anticoagulant are the most common. They are also associated with recurrent fetal loss. These antibodies are also found in association with thrombosis in the multistystem disorder Systemic Lupus Erythematosus. Antinuclear antibodies are the most common autoantibodies found in this disorder\(^{(19)}\).

The mechanism by which antiphospholipid antibodies induce a hypercoagulable state is not fully understood. They have been shown to inhibit activated protein C anticoagulant activity, and impair fibrinolysis\(^{(20)}\). Ieko M and team demonstrated this by examining the effects of monoclonal anticardiolipin (aCL) antibodies and beta2-glycoprotein I (beta2-GPI), which is required for formation of the aCL epitopes, on activated protein C (APC) and on fibrinolytic activity.\(^{(20)}\) Activated protein C inhibits blood coagulation by proteolytic degradation of factors VIIIa and Va. Activated protein C resistance is caused by defective factor V resulting from a single missense mutation in the factor V gene. The mutated factor Va expresses normal factor V procoagulants activity but is less sensitive to activated protein C-mediated degradation. This leads to stabilization of the prothrombinase complex, increased rate of thrombin generation, and feedback activation of factors VIII and V. The increased rate of activation of the coagulation cascade, concomitant with loss of factor V-
dependent activated protein C cofactor activity, potentiates the activated protein C resistance and results in a hypercoagulable state.

Antibody positive patients with venous thromboembolism have a high risk of recurrence. Knowledge of antiphospholipid antibody status is therefore important in assessing future risks, and the duration of anticoagulation.

**HYPERHOMOCYSTEINAEMIA: both an inherited and acquired thrombophilia**

High levels of homocysteine result in endothelial damage, and are associated with venous and arterial thrombosis\(^{(21)}\).

Hyperhomocysteinaemia may result from genetic or nutritional deficiencies, or a combination of both. The most common genetic abnormality is the thermolabile variant of the methylene tetrahydrofolate reductase (MTHFR) enzyme, involving a C to T substitution at position 677. Homozygotes have a prevalence of 5-30% in European populations. Again considerable heterogeneity exists between different populations, and this mutation appears to be rare amongst Black South Africans\(^{(7)}\).

Hyperhomocysteinaemia may also result from vitamin deficiencies; vitamin B12 is a cofactor for this enzyme, and folate a co-substrate. Homocysteine levels are inversely correlated with folate concentration, and adequate folate can compensate for poor enzyme activity due to MTHFR abnormality. Most European studies of patients have not shown an increased risk of VTE due to this enzyme mutation\(^{(22)}\). However, communities with a high level of nutritional
deprivation may be more at risk. In South Africa, hyperhomocysteinaemia is common in women with pre-eclampsia, despite a low frequency of MTHFR deficiency\(^{23}\). Hyperhomocysteinemia may be more common and a stronger risk factor for VTE in developing countries due to a high prevalence of vitamin deficiency.

**THE HAEMOSTATIC SYSTEM IN PREGNANCY**

Pregnancy is a hypercoagulable state. The procoagulant system is enhanced and there is decreased anticoagulant activity. Pregnancy may be regarded as an acquired form of thrombophilia.

Factors V, VII, VIII, X and fibrinogen increase in pregnancy\(^{24}\). Factor VIII is elevated from 15-16 weeks, reaching a peak at 36- 40 weeks that is sustained for the first 3 days postpartum. Mean concentrations from the third trimester onwards are greater than 175 IU/dl, peaking at over 200 IU/dl. A wide range of factor VIII levels is found in individual women; the maximum level recorded was 570 IU/dl, compared to a maximum of 193 IU/dl six weeks postpartum. Recent research on non-pregnant patients suggests that factor VIII levels that are normal for pregnancy are associated with an increased risk of VTE in non-pregnant patients\(^{15,16}\).

Fibrinogen concentrations also rise during the third trimester, and peak in the early postpartum period, at 4.61g/L. None of the mean values were above 5.0 g/L, however the highest individual value was over 6.0 IU/L from 31 weeks onwards. Factor XI and factor XIII levels have not been analysed in pregnancy\(^{24}\).
While clotting factors are increased, anticoagulant activity falls. Protein S levels are lower in pregnancy, antithrombin falls slightly, but there is no overall change in protein C concentration\(^{(24)}\).

However, activated protein C resistance is enhanced, due to the increased levels of factors V and VIII, and decreased levels of protein S\(^{(22)}\). VTE can occur at any time in pregnancy, and fatal events early in the first trimester are documented. The early puerperium is considered the time of highest risk\(^{(25)}\). Mechanical risk factors related to pregnancy are important; pressure on the pelvic veins during late pregnancy, damage to pelvic vessels at delivery, and immobility following caesarean section. However the majority of thrombotic events associated with pregnancy occur in the antenatal period\(^{(22)}\). Women with an underlying genetic thrombophilia are most at risk of VTE during pregnancy. At least 50\% of European women with a new episode of venous thrombosis in pregnancy have a thrombophilic disorder\(^{(26)}\).

The risk of venous thromboembolism for pregnant women with inherited thrombophilia is difficult to ascertain with certainty. All the available data relates to populations of European origin. Similarly to non-pregnant patients, antithrombin deficiency is the most thrombogenic with a reported incidence of pregnancy-associated VTE between 12-60\%\(^{(22)}\). Risks increase when more than one mutation is present; the combination of the factor V Leiden mutation and the G20210A prothrombin gene mutation results in an odds ratio of 107 for VTE compared to 9.3 and 15.2 respectively for these mutations individually\(^{(27)}\).
OBJECTIVE OF THIS STUDY

The specific objective of this study was to determine the prevalence of known genetic and acquired risk factors underlying venous thromboembolism in Black and Coloured pregnant women in Cape Town South Africa, as well as to determine the incidence of venous thromboembolism in the same population.

STUDY DESIGN

A prospective observational study.

STUDY CENTRE

This study took place at Groote Schuur Hospital Maternity Centre, Cape Town which is a tertiary referral centre for the Peninsula Maternity and Neonatal Service (PMNS). All women booked within the Peninsula with a history of venous thromboembolism or a suspected new episode were referred. Ethics committee approval was obtained prior to commencing the study.

Written informed consent was obtained from all patients. Blood was taken for thrombophilia testing at the time of first presentation. All patients were fully informed about their results, and the implications discussed. All women were given a written record of their results, so that these are available should they ever present for medical or obstetric care at another institution. We also asked all women for contact addresses and telephone numbers, so that if additional tests for VTE become available in the future, they can be invited back for screening.
All necessary care was provided in the High Risk Antenatal clinic at Groote Schuur Hospital.

INCLUSION CRITERIA

1. Pregnant women with new onset VTE.
2. Women at high risk for VTE in pregnancy.

This included the following groups:

- Women with history of VTE, whether in pregnancy or related to pregnancy. A confirmed prior event was defined as one for which ongoing anticoagulation had been given for at least 3 months.
- Women who were already pregnant at referral or who had a history of previous thromboembolism and were planning to be pregnant were recruited.
- Women with one or more first degree relatives who had a confirmed episode of VTE. Where possible this was verified with case notes; although in the absence of objective data, a confirmed episode was defined as one for which anticoagulation was given for at least 3 months. The above inclusion criteria was targeted through-out the study but unfortunately due to several factors we only managed to recruit pregnant women who fulfilled the above inclusion criteria, hence all the patients were pregnant women.

Secondary obstetrics hospitals (New Somerset and Mowbray Maternity Hospitals) and Midwife Obstetrics Units and local casualty departments were
requested to refer patients directly to Groote Schuur Hospital.

METHODS

THROMBOPHILIA SCREENING

The following blood tests were performed:

**Anticoagulant activity** -

1. Antithrombin concentration
2. Protein C concentration
3. Protein S concentration
4. Activated protein C resistance
5. Factor V Leiden mutation

**Procoagulant activity** -

1. Prothrombin 20201A mutation
2. Factor VIII concentration
3. Fibrinogen

**Antiphospholipid antibodies** -

1. Anticardiolipin antibodies-repeated 6 weeks apart if initial testing was positive.

**Hyperhomocysteinaemia** -

1. Fasting and post methionine load homocysteine levels
2 Methylene tetrahydrofolate reductase genotype

Women were asked to attend the antenatal clinic having fasted from midnight, and blood taken for fasting homocysteine levels and the other tests as listed above. They were given methionine at 100mg/kg. A post load homocysteine level were taken 6 hours later, with only low methionine foods consumed prior to this time. Women were given sandwiches after the post load homocysteine. Even though methionine loading in pregnancy is controversial, it was the opinion of the study team at that time that metionine loading in our study group was essential in obtaining the right results.

LABORATORY METHODS

Tests for anticoagulant and procoagulant activity were performed in the Haematology Laboratory. Genetic testing was performed in the molecular biology laboratory of the Department of Haematology. Standard laboratory methods were used.

CLINICAL MANAGEMENT

1. Women with suspected VTE in the index pregnancy

Women presenting with suspected VTE had blood taken for investigation prior to receiving any anticoagulation.

A full history and examination was obtained. If VTE could not be excluded on clinical grounds, anticoagulation with intravenous heparin was commenced. Women with suspected deep vein thrombosis had duplex doppler ultrasound
examination with venography being carried out when that was negative. Women with suspected pulmonary embolism had ventilation/perfusion scanning with a spiral computed tomography being performed in cases of diagnostic uncertainty. We know that d-dimer testing has an excellent negative predictive value, but the author was not aware of any study in pregnancy which showed a reliable correlation between d-dimer levels and risk of thromboembolism. Hence d-dimer testing was not done.

Long-term anticoagulation was given to women in whom testing was definitive, or where there was a high probability of VTE. In equivocal cases, the decision rested with the consultant in overall charge of the patient. Confirmed VTE was defined as any event requiring long-term anticoagulation. All blood specimens were frozen and stored after venepuncture. Samples from women with confirmed VTE were subsequently analysed.

All women with confirmed VTE received their antenatal and intrapartum care at Groote Schuur Hospital, and were seen for a follow up appointment 6 weeks postpartum.

2. Women with a personal history of VTE

A full history of any previous events was obtained, including the use of anticoagulants, and any investigations carried out for thrombophilia. If possible, previous case notes were obtained. Details of other predisposing factors were sought including previous pregnancies, use of the combined oral contraceptive pill, and prolonged periods of immobility. A thorough family history was taken. Blood was taken for thrombophilia screening at the earliest possible opportunity.

Thromboprophylaxis was prescribed where necessary by the obstetric team
managing the patient. All women with a previous VTE received anticoagulation during the intrapartum period and for 6 weeks postnatally. Thromboprophylaxis were in the form of either low dose aspirin, low molecular weight heparin or heparin and TED stockings, or a combination of the above as was deemed clinically appropriate by the team involved in the day to day care of the patient.

3. **Women with a family history but no personal history of VTE**

Women were referred to Groote Schuur Hospital for assessment and venepuncture. A full family history was taken, and full information sought about the nature of the event, the type and duration of treatment, and any prior investigation for underlying thrombophilia. If possible, case notes were obtained for family members with their permission.

Women found to have an underlying thrombophilia received further antenatal and intrapartum care at Groote Schuur Hospital. They were given thromboprophylaxis in labour and for 6 weeks postpartum. Thromboprophylaxis were mainly in the form of TED stockings and low molecular weight heparin as well as heparin. Post partum, most patients were converted to warfarin.

**SAMPLE SIZE**

The incidence of VTE in pregnant women in South Africa is unknown. In European populations, symptomatic VTE occurs in approximately 0.65 per thousand pregnancies\(^{(22)}\). Approximately 30,000 women book within the Peninsula Maternity Service each year. On this basis we expected to see at least 20 women per year with new presentation of VTE. We expected to test
30-50 women per year with a previous history of VTE. The number of women referred for investigation only on the basis of a family history is more difficult to estimate.

STATISTICAL ANALYSIS

Continuous data were compared using the student t-test and appropriate non-parametric statistics used for data that were not normally distributed. Categorical data were compared using the Chi-squared test.

RESULTS

During the study period of August 2000 to August 2002 there were 56338 deliveries in the PMNS. The total number of patients recruited for the study were 58. There were no white patients. Four (6.9%) of the patients were eliminated from the study because the history was inconclusive. At the time of writing, 5 (9.3%) patients obstetric notes could not be traced for further data analysis, all these patients had been entered on the basis of a proven new VTE in the index pregnancy. It is therefore appropriate to include these 5 patients in calculating for the incidence of VTE in the PMNS since they were to have VTE in the index pregnancy. These 5 patients were excluded from further data analysis since we could not obtain further information because of the missing folders. The data analysis excluding the calculation for the incidence of VTE, is therefore based on 49 patients of whom 26(53.1%) had VTE during the index pregnancy and 23 (46.9%) had either a family history or a previous history of VTE or thrombophilia. Seventeen (34.7%) of these patients were of Black race and 32(65.3%) were of coloured race. The prevalence of thrombophilia detected
during the study is shown in table I. The incidence of VTE in the PMNS per 1000 deliveries was 0.55, as calculated on the basis of 31 patients with proven new VTE during the index pregnancy.
TABLE I -

PREVALENCE OF KNOWN THROMBOPHILIA IN PMNS

<table>
<thead>
<tr>
<th>FACTOR</th>
<th>NORMAL VALUES</th>
<th>PREV VTE</th>
<th>NEW EVENT</th>
<th>PREV/PN</th>
<th>TOTAL (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FIBRIN</td>
<td>2-5g/L</td>
<td>2</td>
<td>13</td>
<td>0</td>
<td>15(30.6)</td>
</tr>
<tr>
<td>FVIII</td>
<td>50-200%</td>
<td>5</td>
<td>13</td>
<td>1</td>
<td>19(38.7)</td>
</tr>
<tr>
<td>PROT.S</td>
<td>&gt;26%</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1(2.04)</td>
</tr>
<tr>
<td>MTHFR</td>
<td>2(het)</td>
<td>7(het)</td>
<td>1(het)</td>
<td>0</td>
<td>10(20.4)</td>
</tr>
<tr>
<td>ACA</td>
<td>&gt;10</td>
<td>6</td>
<td>9</td>
<td>2</td>
<td>17(34.6)</td>
</tr>
<tr>
<td>PROT.C</td>
<td>60-140%</td>
<td>2</td>
<td>3</td>
<td>0</td>
<td>5(10.2)</td>
</tr>
<tr>
<td>ATIII</td>
<td>80-120%</td>
<td>2</td>
<td>6</td>
<td>1</td>
<td>9(18.37)</td>
</tr>
<tr>
<td>APCR</td>
<td>2-4.5</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>4(8.16)</td>
</tr>
<tr>
<td>F5LEID</td>
<td></td>
<td>0</td>
<td>1(het)</td>
<td>0</td>
<td>1(2.04)</td>
</tr>
<tr>
<td>PROTHR</td>
<td>1(het)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1(2.04)</td>
</tr>
<tr>
<td>HOMOC</td>
<td>&lt;13.7μmol/L</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>1(2.04)</td>
</tr>
</tbody>
</table>

Codes for Table I, II, III and IV -

1. GSH = Groote Schuur Hospital
2. PREV = Previous
3. VTE = Venous Thromboembolism
4. PN = Post Natal
5. FIBRIN= Fibrinogen
6. FVIII = Factor VIII
7. PROT.S = Protein S
8. MTHFR = Methylene tetrahydrofolate reductase
9. ACA = Anti Cardiolipin Antibody
10. PROT.C = Protein C
11. ATIII = Antithrombin III
12. APCR = Activated Protein C Resistance
13. F5 LEID = Factor V Leiden
14. PROTHR = Prothrombin
15. HOMOC = Homocysteine
16. GEN. POP = General population
17. PREG = Pregnancy  
18. Het = Heterozygous  
19. Hom = Homozygous  
20. PREV/PN = History of previous posnatal VTE  
• The number of cases indicated in each column in tables I, II,III and IV are the number of cases found with thrombophilia values above the normal in pregnancy

**TABLE II -**

**COMPARISON OF EUROPEAN AND PMNS PREVALENCE**

<table>
<thead>
<tr>
<th>FACTOR</th>
<th>VTE IN PREGNANCY (PMNS)</th>
<th>VTE IN PREG (EUROPEAN)</th>
<th>GEN. POP (EUROPEAN)</th>
</tr>
</thead>
<tbody>
<tr>
<td>VTE/1000</td>
<td>0.6</td>
<td>0.5-1.0</td>
<td></td>
</tr>
<tr>
<td>FIBRIN</td>
<td>30.6%</td>
<td>No data</td>
<td>3%</td>
</tr>
<tr>
<td>FVIII</td>
<td>38.7%</td>
<td>No data</td>
<td>19%</td>
</tr>
<tr>
<td>PROT.S</td>
<td>2.0%</td>
<td>7.17%</td>
<td>0.2-0.4%</td>
</tr>
<tr>
<td>MTHFR</td>
<td>20.4%(Het.)</td>
<td>9.6%</td>
<td>5-30%(Hom.) 0.3-1.4%(Het.)</td>
</tr>
<tr>
<td>ACA</td>
<td>34.6%</td>
<td>No data</td>
<td></td>
</tr>
<tr>
<td>PROT.C</td>
<td>10.2%</td>
<td>7-17%</td>
<td>0.2-0.4%</td>
</tr>
<tr>
<td>ATIII</td>
<td>18.4%</td>
<td>25%</td>
<td>0.1%</td>
</tr>
<tr>
<td>APCR</td>
<td>8.2%</td>
<td>60%</td>
<td>2-.7%</td>
</tr>
<tr>
<td>F5 LEID</td>
<td>2.0%</td>
<td>40%</td>
<td>5.9%</td>
</tr>
<tr>
<td>PROTHR</td>
<td>2.0%</td>
<td>16.9%</td>
<td>2.3%</td>
</tr>
<tr>
<td>HOMOC</td>
<td>2.0%</td>
<td></td>
<td>10%</td>
</tr>
</tbody>
</table>
TABLE III -

RISK FACTOR AND PROBABILITY OF DEVELOPING VTE IN INDEX PREGNANCY

<table>
<thead>
<tr>
<th>RISK FACTOR</th>
<th>NEW VTE IN INDEX PREG</th>
<th>HISTORY OF VTE (personal/family)</th>
<th>P-VALUE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Multiple risk</td>
<td>18/26</td>
<td>8/23</td>
<td>0.02</td>
</tr>
<tr>
<td>&gt;35 yrs</td>
<td>4/26</td>
<td>5/23</td>
<td>0.72</td>
</tr>
<tr>
<td>&lt;35 yrs</td>
<td>22/26</td>
<td>18/23</td>
<td>0.72</td>
</tr>
<tr>
<td>&gt;2 parity</td>
<td>10/26</td>
<td>11/23</td>
<td>0.57</td>
</tr>
<tr>
<td>&lt;2 parity</td>
<td>16/26</td>
<td>11/23</td>
<td>0.40</td>
</tr>
<tr>
<td>Black race</td>
<td>10/26</td>
<td>7/23</td>
<td>0.76</td>
</tr>
<tr>
<td>Coloured race</td>
<td>16/26</td>
<td>16/23</td>
<td>0.76</td>
</tr>
<tr>
<td>&gt;85kg</td>
<td>2/26</td>
<td>4/23</td>
<td>0.40</td>
</tr>
<tr>
<td>&lt;85kg</td>
<td>24/26</td>
<td>19/23</td>
<td>0.24</td>
</tr>
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</table>

TABLE IV -

RACIAL DIFFERENCE AND PREVALENCE OF THROMBOPHILIA

<table>
<thead>
<tr>
<th>FACTOR</th>
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<tr>
<td>Fibrinogen</td>
<td>12</td>
<td>3</td>
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<tr>
<td>FVIII</td>
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<td>7</td>
</tr>
<tr>
<td>Protein S</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Protein C</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>ATIII</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>APCR</td>
<td>3</td>
<td>1</td>
</tr>
<tr>
<td>MTHFR</td>
<td>8</td>
<td>2</td>
</tr>
<tr>
<td>F5 Leiden</td>
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<td>0</td>
</tr>
<tr>
<td>Prothrombin</td>
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<td>0</td>
</tr>
<tr>
<td>Homocysteine</td>
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</tr>
<tr>
<td>ACA</td>
<td>10</td>
<td>7</td>
</tr>
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</table>
RESULTS OF PATIENTS WITH MULTIPLE THROMBOPHILIA RISK

The total number of patients with multiple thrombophilias was twenty-six (53.31%). Sixteen (32.6%) had two thrombophilias. Six (12.2%) had three thrombophilias and four (8.2%) had four thrombophilias.

RESULTS OF MATERNAL DATA

The median maternal age was 26 years with a range of 15-42 years. The median gravidity was 2.8 with a range of 1-7. The median parity was 2.6 with range of 0-5. The average maternal weight in kilograms was 64.1 with a range of 52-120. The maternal height in metres ranged from 1.4-1.80.

RESULTS OF MATERNAL OUTCOME

There were no maternal deaths from the study patients during the study period. One patient developed pulmonary embolism during the pregnancy. This patient was included on the basis of a previous arterial thrombosis and had combined protein S and protein C deficiency. One patient developed bilateral deep vein thrombosis (DVT) during labour. She was found to be heterozygous for MTHFR. Another patient required amputation of the left leg post partum as a result of compartment syndrome. Her thrombophilia screen confirmed protein C and protein S deficiency. One patient developed abruptio-placentae which was not associated with hypertension. Her thrombophilia screen was negative. There was only 1 patient with a recurrent VTE. Her thrombophilia screen was negative. She weighed 120 kilograms and her parity was 5.
RESULTS OF DELIVERY DETAILS
The average gestation at the time of delivery was 35.5 weeks. Eighteen (36.7%) patients went into spontaneous labour. Thirteen (26.5%) patients had labour induced, 24 (48.97%) patients had a caesarean delivery, half of which were emergency caesarean sections. There were 2(4.08%) termination of pregnancies (one at 13 weeks for very pre-term pre-labour rupture of membranes, and the other due to severe early onset pre-eclampsia at 24 weeks). The average estimated blood loss at delivery was 323 ml with a range of 100-600ml. The average estimated blood loss at caesarean section was 400ml and that during a vaginal delivery was 200ml. The majority of these patients (78%) had regional anaesthesia for caesarean section with no reported anaesthetic complications.

RESULTS OF NEONATAL OUTCOME
The average birth weight was 2560g with a range of 1100-3620g. There were four sets of twins. There were two macerated still-births. Intra-uterine death were diagnosed antenatally. The cause of still-birth was unknown in both cases despite a full workup including postmortem studies. The thrombophilia screen of the above two mothers were that of protein S deficiencies. Five babies required neonatal high care. The median number of days of neonatal high care was 5 days with a range of 1-64 days. Six babies required kangaroo mother care for a duration ranging from 1-20 days. There was one neonatal death from complications of pre-maturity at 28 weeks of gestation. The calculated perinatal mortality rate (PNMR) is 53.57 per 1000 deliveries. The average PNMR for the PNMS is 30 per 1000 deliveries. PNMR is calculated based on fetal viability gestation of 28 weeks. Two babies had nasal hypoplasia, the mothers of both children having entered the study already taking warfarin. There were two babies
with neonatal jaundice that required photo-therapy. There were 3 babies with hyaline membrane diseases.

DISCUSSION

The incidence of VTE in the PMNS of 0.55 per 1000 deliveries is similar to that of the European incidence between 0.5-1.0 per 1000 deliveries. The true incidence in the PMNS might be higher since we only included patients who had conclusive evidence of VTE.

The prevalence of inherited thrombophilia in the PMNS appears to be lower than among the European population although the data are too few to sustain any conclusions.

A number of factors may influence the interpretation of the data presented including:

a) Physiological variation with increasing gestational age. In our study, blood was taken once and not serially with increasing gestational age. Blood was also taken at different gestational ages at the time of presentation and therefore the values cannot be stratified according to gestation and compared to European studies. The physiological ranges used in our study are based on normal ranges for non pregnant patients and because the study was uncontrolled it is not possible to define what values should be interpreted as abnormal in pregnancy. To answer this question, we propose a controlled larger study in the obstetric population to establish a reference range. Arising from these considerations, there are several known physiological changes in pregnancy that would influence the concentrations of clotting factors. For example, during pregnancy there is a fall in protein S levels during the second trimester that could lead to a
misdiagnosis of protein S deficiency. It is therefore essential to know the normal
pregnancy range before diagnosing protein S deficiency. In the absence of clearly
defined pregnancy reference ranges, we chose to use an upper limit of 5g/L to
define hyperfibrinogenenaemia since values greater than 5g/L are associated with a
3-4 fold increased risk of VTE in non-pregnant patients. Defined in this way, the
prevalence of 30.6% was much higher than that found in the European
studies\(^{(18,24)}\)
b) Acute phase response and changes associated with VTE: Acute events,
including thromboembolism may induce changes in clotting factor concentration.
For example, if extensive venous thrombosis develops followed by investigation
of the coagulation profile, antithrombin levels may be reduced giving a false
impression of antithrombin deficiency. The prevalence of 18.37% in this study
was much lower than the European findings\(^{(18,24)}\) despite the fact that 53.4% of
our patients had acute VTE in the index pregnancy. The data may be further
contained by qualitative abnormalities. There are two classes of ATIII
deficiency. The most common is type I ATIII deficiency which is characterized
by both reduction in ATIII concentration and activity. Type II ATIII deficiency is
associated with normal ATIII concentration but decreased activity. In our study
the different types of ATIII were not differentiated by the laboratory.
c) The effects of treatment: the concentration of clotting factors may be affected
by anticoagulation. Heparin induces a decline in ATIII levels and Warfarin
decreases protein C and protein S levels. In our study, testing was deferred to the
post-partum period if patients presented on treatment.
d) The influence of multiple risk factors: In patients with multiple risks (ie two or
more thrombophilias) there was a significantly higher number who had VTE in
the index pregnancy compared to those who did not. This finding agrees with
other studies\(^{(4,25)}\). Other factors associated with VTE such as age, weight and race did not correlate with increased risk. This finding is in contrast to the findings published in the European literature\(^{(4,22,25,27)}\) and probably reflects the small numbers in this study.

This study agrees with the findings of Rubinstein et al\(^{(6)}\) as well as other South African studies\(^{(8,9)}\) with respect to the very low prevalence of factor V Leiden mutation in the population. We therefore recommend not to screen for factor V Leiden mutation in black obstetric patients with VTE except in few selected cases where there is a known family history of factor V Leiden or in cases where all screening tests are negative and there are no acquired risk factors to explain the presence of VTE in a particular patient.

Maternal morbidity in this study was low. There was only one patient who had a recurrence of VTE. This patient did not have genetic thrombophilia but rather two acquired risk factors of increased weight (she was the heaviest patient) and increased parity. This finding is in agreement with the European studies\(^{(4,25)}\) where a low recurrence rate was found. The mode of delivery in these patients shows a very high caesarean section rate of 48.9% as opposed to an overall caesarean section rate within the PMNS of 17.1%. The indications for caesarean section varied widely and were similar to the indications for caesarean section in the general obstetric patients at GSH. The reason for such a higher caesarean section rate in this study cannot be explained although the link between thrombophilia and adverse pregnancy outcome would need to be explored. The assessment of blood loss at delivery (both vaginal and caesarean delivery) is no higher than the average blood loss in the general population. This finding is also in agreement with published literature and increased blood loss as a result of anticoagulation is
unlikely. Anaesthetic complications from regional anaesthesia as a result of anticoagulation were not apparent either immediately or in the long term. This is consistent with previous work concerning the safety of regional anaesthesia in anticoagulated patients when due precautions are taken.

The neonatal outcome was not as favourable as the maternal outcome. The perinatal mortality rate (PNMR) was 53.57 per 1000 deliveries which was much higher than the PMNS average of 30.0 per 1000 deliveries. There was one neonatal death as a result of complications of prematurity (hyaline membrane disease) at 28 weeks of gestation. The cause of preterm labour was idiopathic. This is difficult to attribute neonatal death directly to the maternal risk of thrombophilia as well as to maternal anticoagulation. The two stillborn babies were macerated and the cause of the stillbirths was unexplained despite full investigations to detect underlying causes. There was also a high neonatal morbidity observed in this study. The two babies with nasal hypoplasia were linked to maternal use of Warfarin during pregnancy. Five (8.9%) of these babies spent a substantial amount of time (average 11.2 days) in neonatal high care.

CONCLUSION

Just over two decades ago thrombophilia as a cause for venous thromboembolism in pregnancy and the puerperium was not recognised as a problem. The only known major thrombophilic disorder was a deficiency of antithrombin. In the early 1990’s as few as 10-15% of patients with deep vein thrombosis had identifiable genetic risk factors. Currently 50-70% of patients with VTE in pregnancy or the puerpium will have identifiable risk factors. Clinical thrombosis is now considered a multi-causal disease, resulting from interaction between genetic and acquired risk factors. The
presence of thrombophilia alone does not necessarily lead to a clinical event and it is impossible to predict which patients will develop VTE. Thrombophilia screening as a way of preventing adverse pregnancy outcome is more controversial. Anticoagulation is costly and potentially harmful and should be based on randomised controlled studies. Prophylaxis against VTE is warranted for pregnant women with thrombophilia who have had a previous VTE event and the Royal college of Obstetrics and Gynaecology guideline on thromboembolic disease in pregnancy and the puerperium suggests that these patients should be treated\(^{(35)}\). Future studies must be controlled and other thrombophilic risk factors such as the sticky platelet syndrome, factor XI and factor XIII in the local obstetric population investigated.
REFERENCES


Hyperhomocysteinaemia is common amongst South African women with Pre-eclampsia (in preparation)


Lancet 1999; 353: 1258- 1265


