Coagulation Profile According to Gestational Age in Pregnant Nigerian Women

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Abstract

Background: This study was carried out to highlight the reversible physiological changes in pregnancy with demonstrable hypercoagulability though counterbalanced by increased fibrinolytic activity. An imbalance in these physiological changes may result in thrombotic disorder or haemorrhage.

Method: Coagulation tests ((PT, PTT_k, fibrinogen level and factor $VIII^C$) were performed on 150 healthy pregnant Nigerian women, compared with 50 non-pregnant healthy Nigerian women using standard methods.
Results: The outcome of coagulation screening tests consistently showed shortening of PT and PTTk in each trimester of pregnancy while the factor $VIII^C$ and fibrinogen level increased with each trimester.
Conclusion: It is therefore suggested that serial coagulation studies should be performed on all pregnant women, particularly where there is an additional predisposing factor to DVT such as inherited thrombophilias and any of the hypercoagulable states. Where global tests of coagulation and specific factors assay suggest moderately severe hypercoagulable state, prophylactic anticoagulation should be considered.

Key Words: Pregnancy, Hypercoagulability, Fibrinolytic activity, Coagulation Screening and factor assay

Introduction

Normal haemostasis is a complicated phenomenon during which the vascular endothelium, platelets, procoagulant factors, coagulation inhibitors and the fibrinolytic system interplay to keep a balance between thrombosis and haemorrhage. Normal pregnancy is accompanied by significant alteration in all aspects of Virchows triad: venous stasis, endothelial damage, enhanced coagulation, ultimately shifting the equilibrium towards a prothrombotic state [1]. Placental separation is an acute and severe challenge to haemostasis [2] hence these changes serve to ensure haemostasis at the placental bed and prevent severe bleeding during delivery and the puerperium. Ongoing intra and extra vascular fibrin formation in the utero-placental vasculature is a major contributor to the haemostatic changes in pregnancy.

However major changes in coagulation and fibrinolytic system may result in both thromboembolic disorders and haemostatic failure [3]. Pregnancy is associated with a hypercoagulable state, [4] resulting in increased thrombotic risk during the antenatal period, labour, and in postnatal period [5]. The risk is escalated in individuals with inherited thrombophilia like protein C and S deficiency, factor V leiden, hereditary ATIII deficiency, factor II G20210A deficiency. Acquired causes of thrombophilia/systemic diseases like antiphospholipid syndrome, nephrotic syndrome, Paroxysmal Nocturnal Haemoglobinuria (PNH), cancer, sepsis, inflammatory bowel disease, myeloproliferative disorders etc; heighten the risk of thrombosis [6]. Efficient uteroplacental circulation determines the successful outcome of pregnancy. A bleeding tendency would occur in pregnancy when there is a deficiency of clotting factors, inhibition of the clotting process, or excessive activity of fibrinolytic system [7].

Studies indicating the coagulation profile at each trimester are lacking in this environment. Therefore, this study was carried out to find out the baseline value of PT, PTT_k and to determine correlation between fibrinogen, factor $VIII^C$ and gestational age. This study also aimed at providing standard values for PT, PTT_k, Fibrinogen and factor $VIII^C$ in Nigerian pregnant women.

Materials and Methods.

One hundred and fifty healthy pregnant women attending the antenatal clinic of university College Hospital, Ibadan; aged 19 to 35 years, without a history of coagulation disorder and who were not on any drug that could affect coagulation within the past six months, were recruited into this study. Fifty (50) non-pregnant women, age and gestational-age matched were selected as controls.
Samples for coagulation tests were taken as follows:

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4.5ml of blood was obtained from each of the study and control subjects and added to 0.5ml of 3.2% trisodium citrate. The citrated samples were centrifuged for 15mins at 2,000rpm to obtain platelet poor plasma. The plasma from each sample was carefully aspirated using plastic Pasteur pipette and stored at 20^0C prior to analysis.

Prothrombin Time was performed using the standard method of Quick A.J 1942 [8]. Partial Prothrombin Time with Kaolin (PTT_k) was performed according to Macpherson and Hardisty 1961 [9]; Procter and Rapaport 1961 [10]. Fibrinogen estimation was done according to the method of Clauss (1957)[11], Ellis and Stransky (1961)[12]. The one-stage assay for factor $VIII^C$ was done using the method of Pitney (1956) [13]. The principle is based on the partial thromboplastin time with kaolin (PTT_k). It consists of comparing the ability of dilution of the patients plasma and of a standard plasma to correct the PTT_k of a plasma known to lack factor $VIII^C$ but containing all other factors required for normal coagulation.

Fibrinogen estimation was done according to the method of Clauss as follows:- 0.2ml of 1 in 10 dilution of the citrated sample was placed in khan tubes in a water bath at 37^0C; 0.2ml of thrombin solution was added and a stopwatch was started simultaneously. The tube was tilted at intervals and the time for fibrin formation was noted. The tests were done in duplicate and the mean time was taken. The fibrinogen concentration was determined for each sample from the standard curve.

Table 1:

Mean Values of PT, PTT_k & Fibrinogen $FVIII^C$ of Pregnant Women and Controls in all Trimesters

Trimester	PT (Secs.)	PTT_k (Secs.)	Fibrinogen (g/dl)	Factor $VIII^C$ (%)
First	11	35	5.03ÿ3.4	111ÿ1.8
Second	10	33	5.67ÿ5.4	151ÿ2.1
Third	10	32	5.96ÿ6.0	119ÿ0.7
Control	13	36	4.2ÿ2.3	70ÿ7.1

Table 2:

Intra-group Comparison of Mean Fibrinogen Between Control and the Study Subjects in the First Trimester, First *vs.* Second Trimester, Second *vs.* Third Trimester

Groups Compared	X ÿ sd	t value	p value
Control Subjects *vs.* Study Subjects !st Trimester	4.18ÿ2.30 5.30ÿ3.54	1.5	0.001 (S)
1st Trimester Study Subjects *vs.* 2nd Trimester Study Subjects	5.30ÿ3.54 5.68ÿ5.40	0.90	0.001. (S)
2nd Trimester Study Subjects *vs.* 3rd Trimester Study Subjects	5.68ÿ5.40 5.96ÿ4.10	0.30	0.01 (S)

S = Statistical significance

Results

The mean PT, and PTT_k were shorter than that of the controls in all trimesters and shortest in the third trimester.

Table 1 shows the mean PT, PTT_k, Fibrinogen, and factor $VIII^C$ for the control subjects and for each trimester of pregnancy. Fibrinogen levels were higher than in the control values but highest in the third trimester. Factor $VIII^C$ level in each of the trimester was higher than that of controls but highest in the second trimester than the first and third trimester.

Table 2 shows the intra-group comparison of means of fibrinogen value for control versus the study subjects in the first trimester, first versus the second trimester, and second trimester versus the third trimester. The p values showed statistical significant difference in each of the three groups ($p<0.001$, $p<0.001$ and $p<0.01$ respectively)

Table 3 shows the intra-group comparison of means of factor $VIII^C$ value for control versus the study subjects in the first trimester, first versus second trimester, and second trimester versus third trimester. The p values showed statistical significant difference in each of the three groups ($p<0.001$ in all the group comparison).

Table 3:

Intra-group Comparison of Mean Factor $VIII^C$ Between Control and Study Subjects in the First Trimester, First *vs* Second Trimester, Second *vs* Third Trimester.

Groups Compared	X ÿ sd	t value	p value
Control *vs.* 1st Trimester Study Subjects	70ÿ7.1 111ÿ1.8	25.3	0.001 (S)
1st Trimester study subjects *vs.* 2nd Trimester Study Subjects	111ÿ1.8 151ÿ24.8	7.19	0.001. (S)
2nd Trimester study subjects *vs.* 3rd Trimester Study Subjects	151+24.8 119ÿ0.7	8.65	0.001 (S)

S = Statistical significance

Discussion

Pregnancy has been identified as a hypercoagulable state. The levels of markers of haemostatic activation such as fibrinopeptide A and B (prothrombin fragment 1+2), TAT (thrombin antithrombin complex) and D-dimer similar to or higher than those found in patients following thrombo-embolic event, have been observed in pregnancy [14]. Incidence of pregnancy associated VTE is estimated to be 1/1000 deliveries, 5.5-6 times higher than in the female population [15]. The normal haemostatic balance between coagulation factors/coagulation inhibitors, fibrinolytic factors and fibrinolytic inhibitors in pregnancy is tilted towards a hypercoagulable state, which is counterbalanced by increased fibrinolytic activity[14, 16] and decrease in factor XI and monocytes tissue factor expression [17,18]. Substantial changes in procoagulant haemostatic changes in preparation for the haemostatic challenge of pregnancy is followed by increase in FXIII and FII in early pregnancy [19,20] although there is a return to non-pregnant level by the third trimester. On the other hand, there is a consistent increase through out pregnancy of factors XII, X, IX, VIII, VII, vWF, Fibrinogen[10,13]. However, relatively low incidence of VTE occur because of the increase in fibrinolytic activity, brought about by increases in endothelial derived PAI-1 during the later stages of pregnancy and placental derived PAI-2 which increases gradually from early pregnancy [19]. In this study, prothrombin time and partial thromboplatin time with kaolin were shorter than the control values, thereby suggesting a hypercoagulable state. The mean prothrombin time of the pregnant women was significantly shorter than that of the control subjects ($p < 0.001$) Similarly the mean value of the Partial thromboplatin time with kaolin in pregnant women was significantly shorter than that of the control subjects ($p < 0.001$).

The results of fibrinogen concentration of the pregnant women studied revealed a significant increase in fibrinogen level compared to controls. The mean fibrinogen level of the control subjects was 4.18g/dl (the range was between 3.7g/dl 4.5g/dl). These values are higher than the mean fibrinogen level of 2.50g/dl in Caucasians with a range of 1.5 4.0g/dl. The high level of fibrinogen in blacks has been attributed to endemic infections and parasite infestations.

The mean fibrinogen level steadily rose from 5.03g/dl in the first trimester, to 5.68g/dl in the second trimester and finally to 5.96g/dl in the third trimester.. This is in agreement with other studies [21], [22] that showed that fibrinogen level were always on the increase as the pregnancy advances. Also, Kobayashi *et al* [23] found that fibrinogen was an extremely important factor in maintenance of pregnancy and that pregnancy is associated with increased level of fibrinogen, proportional to the gestational age. Also, there was a significant correlation of fibrinogen level with gestational age in both the study subjects. The increase in factor $VIII^C$ level is in agreement with Caires et al [24], who also found progressive increase in the level of Factor $VIII^C$ with gestational age.

Walker *et al* [25] observed that there was an increase in factor $VIII^C$ level reaching its peak during the third trimester. The present work is in agreement in that from the 1st trimester there was an increase in value from 111% to 151% in the second trimester but with a drop to 119% in the third trimester. This findings of an increase in factor $VIII^C$ levels in pregnancy in Nigerian patients is in keeping with the previous study of Noe, D.A 1996[26], showed elevated factor $VIII^C$ in pregnant women.

The progressive hypercoagulability demonstrated in pregnancy is nature's way of preventing excessive bleeding during parturition. This is of tremendous advantage in individuals with inherited factor deficiency like von Willebrands disease (vWD), which tend to improve in pregnancy. vWD is an inherited (usually autosomal dominant) bleeding disorder; characterized by reduced platelet adhesiveness(usually <20% of normal) as a result of quantitative or qualitative defect in the multimeric molecular complex of the glycoprotein subunit (vWF multimers), leading commonly to muco-cutaneous bleeding in spite of normal platelet count[27]

The corollary of the above findings is that increased level of a coagulation factor in pregnancy is not an isolated phenomenon, and that the hypercoagulable state involves a complex mechanism so as to ensure that pregnant women do not bleed to death during childbirth.
During pregnancy and the immediate post partum period, the hypercoagulability could predispose to thrombosis, particularly where there are compounding predisposing factors to deep vein thrombosis (DVT) such as diabetes, prolonged immobilization, pelvic surgery, shock and haemoglobinopathies.

This study is of relevance in clinical practice in that significantly prolonged PTT_k in conjunction with prolonged thrombin time and reduced platelet count; together with hypofibrinogenaemia (which becomes manifest when LDH is greater 240U/ml) in the pre-eclamptic patient; have predictive value in detecting disseminated intravascular coagulopathy (DIC) .[28,29]. The presence of DIC has a positive correlation with increasing severity of Pregnancy Induced Hypertension.[28,29]. In Hadatidiform mole, coagulation profile assessment is necessary to exclude the development of coagulopathy.

In conclusion, this study showed ample evidence of a progressive hypercoagulable state with advancing pregnancy evidenced by shortened PT, PTT_k, progressive increase factor $VIII^C$ and fibrinogen levels compared with the corresponding values in the control subjects. Serial coagulation study is therefore suggested for all pregnant women, particularly in pregnant women with a previous or family history of venous thromboembolic disorders (VTE). Where global tests of coagulation and specific factors assay suggest moderately severe hypercoagulable state, prophylactic anticoagulation should be considered, particularly where there is additional predisposing factor to DVT such as pelvic surgery, shock, or prolonged immobilization. Patients with severe pre-eclampsia and hydatidiform mole may develop coagulopathy and coagulation profile has predictive value in detecting DIC.

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