Hemostatic changes in normal pregnancy

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A B S T R A C T

Normal pregnancy is accompanied by changes in the coagulation and fibrinolytic systems. These include a decrease in platelet count, increases in a number of clotting factors, a decrease in protein S levels, a significant fall in the activity of activated protein C and inhibition of fibrinolysis. These changes may be important for reducing intrapartum blood loss, but they determine an increased risk of thromboembolism during pregnancy and puerperium.

Normal pregnancy is associated with major changes in many aspects of haemostasis all contributing to maintain placental function during pregnancy and to prevent excessive bleeding in delivery. Most changes in blood coagulation and fibrinolysis create a state of hypercoagulability.[1,2] This phenomenon protects the woman from haemorrhage during delivery but predisposes her to thromboembolism both during pregnancy and in puerperium. The changes in the coagulation system in normal pregnancy are consistent with a continuing low-grade process of intravascular coagulation.[3] A summary of the main changes in haemostatic parameters is reported in Table 1.

Platelets

Thrombocytopenia is the most common haemostatic abnormality observed in pregnancy.[1] In many healthy women (around 10%) late pregnancy is associated with thrombocytopenia. At least in part this is due to haemodilution but the increase in mean platelet volume[4] suggests that a compensated state of progressive platelet destruction occurs. Additional evidence of *in vivo* platelet activation in late pregnancy is the increased concentration of β-thromboglobulin[5] and of thromboxane A2 derivatives.[6]

Coagulation system

During pregnancy the concentrations of coagulation factors VII, VIII, IX, X, XII and von Willebrand factor rise significantly, accompanied by a relevant increase in the concentration of plasma fibrinogen.[1,2,7,8] Plasma fibrinogen often increases to over 600 mg/dL in late pregnancy.[9] Factor VII may increase as much as tenfold in pregnancy.[1, 10] The von Willebrand factor and factor VIII are elevated in late pregnancy, when coagulation activity is about twice that in the non-pregnant state.[1] The increase in factor IX concentrations during pregnancy is reported by several authors to be small,[10] as is the decrease in factor XI.[11] After an initial increase, factor XIII falls gradually, reaching 50% of the normal non-pregnant value at term.[12] Factors II and V do not change significantly in pregnancy.[1]

Controversial data have been reported about changes in antithrombin (AT) during pregnancy,[13,14] but AT often remains in the normal range. Protein C activity appears to be unaffected by gestation.[15-18] Protein C antigen levels tend to increase in the second trimester but values are within the normal non-pregnant range.[19] Neutrophil activation is known to trigger endothelial thrombomodulin (TM) proteolysis and to increase TM plasma levels in the third trimester of pregnancy.[20] Other studies found that TM levels increase continually during pregnancy, with a rapid decrease post-partum.[21] Although the fall in free protein S levels in pregnancy is a physiological event, it is unclear whether it contributes to the hypercoagulable state of pregnancy and the increased incidence of thromboembolism. Total protein S has been reported to fall progressively with increasing gestation.[22,23] However, the values for both total protein S and free protein S go below the

Table 1. Main changes in haemostasis factors during pregnancy.

Platelet count	↓
II, V	=
Fibrinogen,VII,VIII,von Willebrand factor,IX,X,XII	↑
XI	=/↓
XIII	↑/↓
Antithrombin	=
Protein C	=/↑
Protein S	↓
Heparin cofactor II	↑
F1+2,TAT,d-dimer	↑
t-PA	↓
ELT,PAI, TAFI	↑

normal range very early.[14] The apparent fall in protein S during the first weeks of pregnancy does not allow a diagnosis of inherited protein S deficiency in pregnant women. Attempts to establish protein S normal levels during pregnancy are not recommended. Heparin cofactor II, another natural coagulation inhibitor, has been reported to increase in plasma during physiological pregnancy.[24]

Protein Z is a vitamin-K-dependent plasma glycoprotein and inhibits the activation of factor X by serving as a cofactor to a plasma proteinase inhibitor. Protein Z deficiency has recently been reported in women with unexplained early fetal losses, and antibodies to protein Z can contribute to adverse pregnancy outcomes. Recent data show a progressive increase in protein Z levels with gestational age in normal pregnancies and a return to normal levels around 6 to 12 weeks postpartum.[25] The normal increase of protein Z during pregnancy may balance the increase of clotting factors to protect pregnant women from thrombosis.[25]

Activated protein C (APC) sensitivity is reduced during pregnancy;[26] at term, 45% of pregnant women have an APC sensitivity ratio below the 5th percentile of the normal range for non-pregnant women of similar age.[27] The reduction in APC ratio is directly related to its value in the non-pregnant state, being most pronounced in the women with the highest APC ratio.[28] About 50% of the healthy women develop APC resistance, which reaches its lowest value by pregnancy second trimester with little further change.[26] This behaviour of the classical APC resistance test has been called 'acquired' APC resistance.[29-31]

In a cross-sectional study no correlation was found between the decrease in the classical APC ratio and the free protein S levels;[29] in another, a negative covariance was found between the APC ratio and FVIII levels in the first trimester[26] and at delivery.[27] Changes in the free protein S concentration are unlikely to contribute significantly to the development of APC resistance during pregnancy. Actually, protein S levels decline progressively during pregnancy and this differs from the pattern of reduction in APC anticoagulant activity, where little change in the APC ratio occurred between the second and third trimesters. In a prospective longitudinal study on healthy pregnant women[19] no correlation was found between the total change in the classical APC ratio and the total changes in FVIII, fibrinogen or protein S.

A modified APC resistance test, which includes sample dilution in FV-deficient plasma prior to the APTT-based assay, has been developed and shown to be useful in screening for factor V Leiden in patients on oral anticoagulants or with antibodies against phospholipids. When this modified APC resistance test was used, the gestation-dependent APC resistance that has been reported with the unmodified test was no longer observable.[32] A significant, gradual increase in the levels of soluble fibrin, thrombin–antithrombin complexes[33] and prothrombin fragment 1+2[34] have been reported. The observed rise in F1+2 between the first and second trimester indicates that a degree of activation of coagulation occurs relatively early in normal pregnancy.[14, 34] Thus an increased thrombin generation is a feature of normal pregnancy.

A concurrent increase in the levels of the fibrinolytic inhibitors plasminogen activator inhibitors 1 and 2 suggests a decrease in fibrinolytic activity. However, the levels of fibrin d-dimer, i.e. fibrin split products, also increases in parallel, so suggesting that fibrinolysis is present.[13]

Fibrinolysis

Plasma fibrinolytic activity is reduced during pregnancy, remains low during labour and delivery, and returns to normal early after placental delivery.[1] Tissue plasminogen activator (t-PA) activity decreases during pregnancy.[35] This is due not only to the gradual increase in plasminogen activator inhibitor-1 (PAI-1), but also

to the increasing levels of plasminogen activator inhibitor-2 (PAI-2).[36,37] PAI-1 values increase during pregnancy and normalize at 5 weeks post-partum.[13]

PAI-2 generally becomes detectable in plasma only in individuals who are pregnant. Because villous cells are the source of PAI-2[38,39] changes in the amount of placental tissue may influence its level in plasma;[33, 40] thus, a positive correlation is found between PAI-2 concentrations and placental weights. The concentration of PAI-2 varies with birth weights[33,40,41] indicating a dependency not only upon the quantity and quality of the placental tissues but also upon fetal growth.

Despite the high levels of PAI-1 and -2, a highly significant positive correlation has been observed between gestational age and d-dimer concentration.[9,42] The increase in D-dimer makes hard the use of this parameter in the exclusion of venous thromboembolism in pregnant patients with clinical suspicion. Attempts have been made to establish specific ranges of D-dimer levels in pregnancy.[9, 43, 44]

In pregnancy, TAFI levels have been reported to remain stable over months and to be correlated only with age in women.[45] No correlation has been found between TAFI and d-dimer levels.[46] More recently a significant increase in TAFI levels[47] and its correlation with the increase in clot lysis time during pregnancy [48] were reported. Moreover, changes in TAFI seem to contribute to the increasing APC ratio of pregnancy.

Changes in coagulation and fibrinolysis after delivery

The increase in clotting activity at the time of delivery is most likely related to expulsion of the placenta and release of thromboplastic substances at the site of separation.[49] In principle, the changes in the haemostatic mechanism during the puerperium were the same as those observed after extensive surgery.[50] The mean platelet count decreases slightly at the time of placental delivery[51] and starts to increase on days 2–5 post-partum. In high-risk patients where thromboprophylaxis is indicated post-partum, the difference in reactive thrombocytosis post-partum, due to operative delivery, ought to be taken into account.

Plasma AT levels significantly rise after normal delivery for at least 2 weeks post-partum. A rise in protein C level has been shown immediately after delivery and still 3 days post-partum.[14,52] The level of total and free protein S increases significantly after delivery from the first day of the puerperium; whereas total protein S normalizes in the first week post-partum, free protein S was reported not normalized at 5 weeks post-partum.[13] At 8 weeks post-partum 15% of the women still have levels below the reference range for non-pregnant women.[19] Thus, the free fraction of protein S does not seem to reach the non-pregnant value within 8 weeks post-partum, which might be taken into consideration when evaluating thrombotic risk.

Both TAT and prothrombin fragment 1+2 levels increase during and immediately after delivery.[53] Three weeks after delivery, blood coagulation and fibrinolysis has generally normalized.[54] The state of compensated, accelerated intravascular coagulation may be necessary for maintenance of the uterine placental interface and preparation for the haemostatic challenge of delivery.[3]

The peak in clotting and platelet activity seems to occur immediately after placental delivery, whereas the peak of fibrinolytic activity is seen during the first 3 hours post-partum,[3] as reflected by an increase in d-dimer levels.

Miscellanea

Microparticles (MP) are membrane vesicles that are shed from various cellular surfaces. There are two mechanisms that can result in microparticle formation, cell activation and apoptosis. MP are associated with thrombotic and inflammatory complications. Endothelial cells produce MP when the cells are exposed to cytokines, such as interleukin-1 and tumor necrosis factor. Circulating platelet microparticle concentration is a marker of platelet activation. Normal pregnancy is characterized by increased levels of platelet and endothelial MP compared to nonpregnant healthy women[55] but the prevalence and the role of MP in gestational vascular complications remain controversial.

Hyperhomocysteinaemia is a strong independent risk factor for venous thromboembolism and is associated with adverse pregnancy outcomes such as preeclampsia, placental abruption, early pregnancy loss and neural-tube defects. Despite a high folate requirement, several studies have reported that homocysteine (Hcy) is lower in normal pregnancy than in the non-pregnant state. Although the exact mechanisms of Hcy lowering during pregnancy are not well understood (even if the physiological increase in glomerular filtration rate plays a role), one possible outcome of lower Hcy may be the protection of women from pregnancy complications and thromboembolism, and thus lower Hcy may contribute to maintaining homeostasis in haemostasis. In a recent study[56] plasma total Hcy was found significantly lower throughout pregnancy compared with nonpregnant controls, with values lowest in the second trimester before increasing toward nonpregnant values in the

third trimester. Importantly, mean total Hcy concentrations were lower in pregnant women taking folic acid supplements than in those not. During the third trimester, total Hcy levels were significantly higher in pregnant women with a history of miscarriage than in women with no previous history.

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