CHAPTER 1

Hematologic Changes in Pregnancy

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Introduction

Normal pregnancy is characterized by profound changes in nearly every organ system to accommodate the demands of the fetoplacental unit. Maternal hematological adaptations to the pregnant state are reviewed in this chapter. The most significant hematological changes are physiologic anemia, neutrophilia, mild thrombocytopenia, increased procoagulant factors, and diminished fibrinolysis.

This chapter will review the pregnancy-associated changes in plasma volume, red blood cells, white blood cells, platelets, and coagulation factors.

Plasma Volume

Plasma volume increases by 10–15% at 6–12 weeks of gestation [1–3], expands rapidly until 30–34 weeks, after which there is only a modest rise. The total gain of plasma volume at term averages 1100–1600 mL and results in a plasma volume of 4700–5200 mL, 30–50% above that found in nonpregnant women [1, 4]. Plasma volume decreases immediately postpartum, then increases again 2–5 days after delivery, possibly because of a simultaneous rise in aldosterone secretion. Plasma volume then decreases; it is still elevated by 10–15% above nonpregnant levels at 3 weeks postpartum, but is usually at normal nonpregnant levels at 6 weeks postpartum.

During pregnancy, plasma renin activity is typically increased and atrial natriuretic peptide levels are slightly reduced, suggesting that the increase in plasma volume represents underfilling due to systemic vasodilatation and the ensuing rise in vascular capacitance, rather than true blood

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volume expansion, which would produce the opposite hormonal profile (low plasma renin activity, elevated atrial natriuretic peptide) [5, 6]. Furthermore, the degree of sodium retention is physiologically regulated, as increasing sodium intake does not produce further volume expansion [7].

Red Blood Cells

Red blood cell mass begins to increase at 8–10 weeks of gestation and steadily rises by 20–30% (250–450 mL) above nonpregnant levels by the end of pregnancy in women receiving iron supplementation [4, 8–11]. Among women not on iron supplements, the red cell mass may only increase by 15–20% [12]. Erythrocyte life span is slightly decreased during normal pregnancy [13].

Erythropoietin levels increase by 50% in normal pregnancies and vary according to the presence of pregnancy complications [14]. The increased plasma erythropoietin induces the rise in red cell mass, which partially supports the higher metabolic requirement for oxygen during pregnancy [15]. Mean corpuscular volume decreases during pregnancy and averages 80–84 fL in the third trimester [16].

Anemia

A greater expansion of plasma volume relative to the increase in hemoglobin mass and erythrocyte volume is responsible for the modest fall in hemoglobin levels (i.e., physiological or dilutional anemia of pregnancy) observed in healthy pregnant women. The greatest disproportion between the rates at which plasma and erythrocytes are added to the maternal circulation occurs during the late second to early third trimester. (Lowest hematocrit is typically measured at 28–36 weeks [16].) Nearer to term, hemoglobin concentration increases due to cessation of plasma expansion and continuing increase in hemoglobin mass. Conversely, the absence of physiologic anemia appears to be a risk factor for stillbirth [17].

Determining a good definition of anemia in pregnant women is not straightforward, given the pregnancy-associated changes in plasma volume and red cell mass, normal differences in hemoglobin concentrations between women and men, ethnic variation between white and black women, and the frequent use of iron supplementation in pregnancy. The Centers for Disease Control and Prevention has defined anemia as hemoglobin levels of less than 11 g/dL (hematocrit less than 33%) in the first and third trimesters and less than 10.5 g/dL (hematocrit less than 32%) in the second trimester [18]. Since hemoglobin and hematocrit

levels are lower in African-American adults, the Institute of Medicine recommends lowering of the hemoglobin cutoff level by 0.8 g/dL in this population [19].

Women with hemoglobin values below these levels can be considered anemic and should undergo a standard evaluation [20]. Sixteen to twenty-nine percent of pregnant women become anemic in the third trimester [21].

Severe anemia with maternal hemoglobin below 6 g/dL has been associated with reduced amniotic fluid volume, fetal cerebral vasodilation, and nonreassuring fetal heart rate patterns [22]. Increased risks of prematurity, spontaneous abortion, low birth weight, growth restriction, and fetal death have also been reported [23]. The administration of lactoferrin to treat iron deficiency anemia in pregnancy requires further investigation. Lactoferrin chelates two ferric ions, decreases interleukin-6, thereby decreasing hepcidin and increasing ferroportin expression.

Iron Requirements

In a typical singleton gestation, maternal iron requirements average close to 1000 mg over the course of pregnancy: approximately 300 mg for the fetus and placenta and approximately 500 mg, if available, for the expansion of the maternal hemoglobin mass. Two hundred milligrams is shed through the gut, urine, and skin. Since most women do not have adequate iron stores to handle the demands of pregnancy, iron is commonly prescribed as part of a prenatal multivitamin or as a separate supplement. In general, women taking iron supplements have a mean hemoglobin concentration that is 1 g/dL greater than that of women not taking supplements. Normal iron indices for pregnancy are listed in Table 1.1.

Folate Requirements

The increase in red cell mass also necessitates an increased folic acid requirement. In nonpregnant women, the daily folic acid requirement is 50–100 mg/d. However, because folate deficiency is associated with neural tube defects (and possibly other birth defects) as well as macrocytic anemia, all women of reproductive age are advised to consume 0.4 mg of folic acid daily [24].

Table 1.1 Normal iron indices during pregnancy

Plasma iron	40–175 μg/dL
Plasma total iron-binding capacity	216–400 μg/dL
Transferrin saturation	16–60%
Serum ferritin	10 μg/dL

Platelet Count

Although platelet counts remain in the normal nonpregnant range in most women during uncomplicated pregnancies [25], mean platelet counts of pregnant women may be slightly lower than in healthy nonpregnant women [26]. Serial platelet counts during uncomplicated pregnancies may [27] or may not [28] decrease, but the mean values in these groups do not necessarily reflect both increases and decreases in individual women [29]. The lower limit of normal platelet counts in pregnancy has been reported to be 106,000–120,000 platelets/μL.

Thrombocytopenia

The most significant obstetrical consideration concerning platelet physiology in pregnancy is thrombocytopenia, which may be related to complications of pregnancy (e.g., severe preeclampsia, HELLP syndrome), medical disorders (e.g., idiopathic thrombocytopenic purpura, thrombotic thrombocytopenic purpura-hemolytic uremic syndrome), or gestational. Gestational or incidental thrombocytopenia is characterized by mild asymptomatic thrombocytopenia occurring in the third trimester in a patient without any history of thrombocytopenia (other than in a prior pregnancy). It is not associated with maternal, fetal, or neonatal sequelae and spontaneously resolves postpartum [30–32]. Platelet counts are typically greater than 70,000/μL.

White Blood Cells

Pregnancy is associated with leukocytosis, primarily related to increased circulation of neutrophils. The neutrophil count begins to increase in the second month of pregnancy and plateaus in the second or third trimester, at which time the total white blood cell counts ranges from 9000 to 15,000 cells/μL [33]. Data from two series reported mean white blood cell counts of 10,000–16,000 cells/μL in laboring patients, with an upper level as high as 29,000 cells/μL [34, 35]; the mean count increased linearly with the duration of elapsed labor [35]. The white blood cell count falls to the normal nonpregnant range by the sixth day postpartum. Dohle bodies (blue staining cytoplasmic inclusions in granulocytes) are a normal finding in pregnant women.

In healthy women with normal pregnancies, there is no change in the absolute lymphocyte count and no significant changes in the relative numbers of T and B lymphocytes [36]. The monocyte count is generally stable; the basophil count may slightly decrease and the eosinophil count may slightly increase. Normal pregnant women can have a small number of myelocytes or metamyelocytes in the peripheral circulation.

Coagulation

Normal pregnancy is a prothrombotic state [37–46]. The circulating levels of several coagulation factors change during pregnancy (Table 1.2):

- Protein S activity and free protein S antigen decrease due to estrogen-induced increases in the complement 4b binding protein and possibly due to other mechanisms related to the hormonal changes of pregnancy.
- Resistance to activated protein C increases in the second and third trimesters.
- Fibrinogen, factors II, VII, VIII, and X increase by 20–200% [47]; von Willebrand factor also increases.
- Activity of the fibrinolytic inhibitors, thrombin activatable fibrinolytic inhibitor (TAFI), PAI-1, and PAI-2 increases [48].
- Factors V and IX remain unchanged and factor XI levels decrease by 30% [47].

The net effect of these changes is to increase the tendency toward thrombus formation, extension, and stability. Normalization of coagulation parameters varies depending on the factor, but all should return to baseline by 8 weeks postpartum.

Table 1.2 Hemostatic changes in pregnancy.

Variables (mean ± SD)	First tri*	Second tri*	Third tri*	Normal range
Platelet ($\times 10^9$ 1^{-1})	275 ± 64	256 ± 49	244 ± 52	150–400
Fibrinogen (g/L)	3.7 ± 0.6	4.4 ± 1.2	5.4 ± 0.8	2.1–4.2
Prothrombin complex (%)	120 ± 27	140 ± 27	130 ± 27	70–30
Antithrombin (U/mL)	1.02 ± 0.10	1.07 ± 0.14	1.07 ± 0.11	0.85–1.25
Protein C (U/mL)	0.92 ± 0.13	1.06 ± 0.17	.94 ± 0.2	0.68–1.25
Protein S, total (U/mL)	0.83 ± 0.11	0.73 ± 0.11	0.77 ± 0.10	0.70–1.70
Protein S, free (U/mL)	0.26 ± 0.07	0.17 ± 0.04	0.14 ± 0.04	0.20–0.50
Soluble fibrin (nmol/L)	9.2 ± 8.6	11.8 ± 7.7	13.4 ± 5.2	<15
Thrombin–antithrombin (μg/L)	3.1 ± 1.4	5.9 ± 2.6	7.1 ± 2.4	<2.7
D-dimers (μg/L)	91 ± 24	128 ± 49	198 ± 59	<80
Plasminogen activator inhibitor-1 (AU/mL)	7.4 ± 4.9	14.9 ± 5.2	37.8 ± 19.4	<15
Plasminogen activator inhibitor-2 (μg/L)	31 ± 14	84 ± 16	160 ± 31	<5
Cardiolipin antibodies positive	2/25	2/25	3/23	0
Protein Z (μg mL^{-1})†	2.01 ± 0.76	1.47 ± 0.45	1.55 ± 0.48	
Protein S (%)†		34.4 ± 11.8	27.5 ± 8.4	

*First tri, 12–15 weeks; second tri, week 24; third tri, week 35.

†First tri, 0–14 weeks; second tri, 14–27 weeks; third tri, 27 weeks or more.

tri, trimester.

Adapted from Bremme [46], table 3, p. 157 and Paidas *et al*. [51], with permission.

Protein S

Protein S (PS) is a vitamin K-dependent glycoprotein with several anticoagulant functions [49]. In the presence of PS, activated protein C inactivates factor Va and factor VIIIa, resulting in reduced thrombin generation. PS also serves as a cofactor for protein C enhancement of fibrinolysis. PS has a direct anticoagulant effect independent of its co-factor function with activated protein C. It prevents the binding of surface phospholipids with factors such as Va, Xa, and VIIIa, thereby decreasing the activation of the factors.

Pregnancy is associated with decreased levels of PS activity and free PS antigen [44, 50]. The significance and degree of decrease in PS levels commonly seen in pregnancy has not been vigorously evaluated. To address this question, we compared second and third trimester PS levels in 51 healthy women with a normal pregnancy outcome with 51 healthy women with a poor pregnancy outcome [51]. Protein S levels were significantly lower in the second and third trimesters among patients with adverse pregnancy outcome compared to patients with normal pregnancy outcome (second trimester 34.4 $\pm$ 11.8% versus 38.9 $\pm$ 10.3%, respectively; and third trimester 27.5 $\pm$ 8.4 versus 31.2 $\pm$ 7.4%, respectively).

Resistance to Activated Protein C

During pregnancy, normal women acquire some degree of resistance to activated protein C (APC), when measured by the first generation global assays and tests that measure endogenous thrombin potential [45, 52, 53].

Factor X

Factor X, its activation to FXa and participation in the activation of prothrombin, is a central element in the generation of thrombin [54]. It is possible that derangements in the control of factor Xa contributes to adverse prothrombotic sequelae in pregnancy.

Protein Z

Protein Z (PZ) is a 62 kDa vitamin K-dependent plasma protein that serves as a co-factor for a PZ-dependent protease inhibitor (ZPI) of Factor Xa [55, 56]. It is a component in the regulation of factor Xa activity in addition to tissue factor pathway inhibitor [57–59]. PZ deficiency increases the prothrombotic phenotype in factor V Leiden patients and has been associated with various adverse clinical sequelae [60–63].

There is a reported increased prevalence of PZ deficiency in patients with unexplained early fetal loss (10–19 weeks of gestation) and other adverse pregnancy outcomes [51, 64–67]. As an example:

- One study reported the odds ratio for fetal loss associated with PZ deficiency was 6.7 (95% CI 3.1–14.8) and noted that the patients with late fetal loss and recurrent miscarriages had lower PZ levels [65].

- Another study found that women with a variety of adverse pregnancy outcomes (e.g., intrauterine growth restriction, preeclampsia, preterm delivery, and antepartum bleeding) had significantly lower PZ levels in each trimester than women with normal pregnancy outcomes [51]. Protein Z levels at the twentieth percentile (1.30 mcg/mL) were associated with an increased risk of adverse pregnancy outcome (OR 4.25, 95% CI 1.5–11.8, sensitivity 93%, specificity 32%).
- An inverse correlation was found between anti-protein Z IgM antibody levels and protein Z concentrations ($p = -0.43$) in patients with recurrent embryonic loss and PZ deficiency [66]. However, the relationship between PZ antibodies and PZ levels is not straightforward. Anti-protein Z IgG antibody and anti-protein Z IgM antibody levels were not correlated with protein Z levels in the entire cohort of patients with normal and abnormal outcomes. The immunological response to coagulation factors in pregnancy requires further inquiry. A recent meta-analysis of 28 case-control studies (33 patient cohorts), including 4,218 patients with thrombotic diseases and 4,778 controls, were analyzed [68]. Low protein Z levels were associated with an increased risk of thrombosis (odds ratio [OR] 2.90, 95% confidence interval [CI] 2.05–4.12; $p < 0.00001$). A significant association was found between low protein Z levels and arterial vascular diseases (OR 2.67, 95% CI 1.60–4.48; $p = 0.0002$), pregnancy complications (OR 4.17, 95% CI 2.31–7.52; $p > 0.00001$), and venous thromboembolic diseases (OR 2.18, 95% CI 1.19–4.00; $p = 0.01$). Thus, protein Z deficiency appears to play a role in thrombotic diseases, including arterial thrombosis, pregnancy complications and venous thromboembolism.

Activation Markers

Activation markers are often increased in pregnancy. Normal pregnancy is associated with both increased thrombin activity, increased soluble fibrin levels (9.2–13.4 nmol/L) and increased thrombin–antithrombin complexes (3.1–7.1 mcg/L), and fibrinolysis, as evidenced by increased levels of fibrin D-dimer (91–198 mcg/L) [69].

Summary and Key Points

The major hematological changes during pregnancy are physiologic anemia, neutrophilia, mild thrombocytopenia, increased procoagulant factors, and diminished fibrinolysis.

- Plasma volume increases by 10–15% at 6–12 weeks of gestation, and then expands rapidly until 30–34 weeks, after which there is only a modest rise.

- Red blood cell mass begins to increase at 8–10 weeks of gestation and steadily rises by 20–30% (250–450 mL) above nonpregnant levels by the end of pregnancy.
- A greater expansion of plasma volume relative to the increase in hemoglobin mass and erythrocyte volume is responsible for the modest fall in hemoglobin levels (i.e., physiological or dilutional anemia of pregnancy) observed in healthy pregnant women.
- The Centers for Disease Control in the United States and Prevention has defined anemia as hemoglobin levels of less than 11 g/dL in the first and third trimesters and less than 10.5 g/dL in the second trimester.
- Mean platelet counts of pregnant women may be slightly lower than in healthy nonpregnant women.
- The neutrophil count begins to increase in the second month of pregnancy and plateaus in the second or third trimester, at which time the total white blood cell counts ranges from 9000 to 15,000 cells/μL.
- There is no change in the absolute lymphocyte count.
- The circulating levels of several coagulation factors change during pregnancy and contribute to the prothrombotic and antifibrinolytic changes associated with pregnancy.

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