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Alteration in blood parameters among normal pregnant women of Abbottabad

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Abstract:
Objectives: *To investigate the hormonal, biochemical, and hematological parameters during three trimesters of normal pregnant women of Abbottabad. To compare the results of the same blood Parameters with the control group of non-pregnant women having similar criteria.*
Materials and Methods: *This is a cross-sectional study conducted between October 2021 and September 2022 at the Women Medical College, Abbottabad in collaboration with Women and Children, hospital (WCH) Abbottabad.*
A total of 90 pregnant women of middle socio-economic class within the age range of 18 to 35 years from the outpatient door (OPD) of WCH were investigated. Twenty-five non-pregnant, non-lactating women of the same age range at days 4-10 of normal menstrual cycle and belonging to the same area and socio-economic background were also included as a control group.
Information from the participants was collected through a pre-designed/ pre-tested questionnaire. The data were analyzed through basic statistical methods.
Results: *The hormones Progesterone, Estradiol, and prolactin were raised ($P< 0.0001$) during pregnancy while Ferritin, Vitamins D3, and B12 declined ($P< 0.0001$) during pregnancy.*
Among Biochemical blood parameters Glucose ($P< 0.019$), Cholesterol ($P< 0.0001$), and Triglycerides ($P< 0.0001$) increased whereas protein ($P< 0.0001$), Albumin ($P< 0.0001$), Urea ($P< 0.002$), Creatinine ($P= 0.5$), iron ($P< 0.0001$), and Calcium ($P=0.3$) decreased during pregnancy.
Hematological parameters like Hemoglobin ($P= 0.0079$). Hematocrit ($P< 0.0001$), red cells ($P< 0.0001$), and platelets ($P= 0.58$) were found low, and Leukocytes ($P=0.029$) and Neutrophils ($P< 0.0001$) were high among pregnant women as compared to non-pregnant women.
Hence Calcium, Creatinine and platelets were significantly ($p>0.05$) not changed, and all other parameters altered significantly ($p<0.05$) after the onset of pregnancy. Besides, the above investigations did not vary with Parity and the other included variables in both the control and study groups.
Conclusion: *Differences were observed in several of the laboratory parameters during pregnancy as compared to the non-pregnant women. The stated changes highlight the need for the establishment of reference ranges specific to pregnancy for every woman's community. Further large-scale studies including more blood parameters to elaborate and verify the facts are needed in different parts of the country regarding the subject.*
Key Words: *Blood parameters, pregnant women, Abbottabad.*

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I. Introduction:

Pregnancy constitutes a special condition in women's life that affects various physiological and metabolic systems. All these effects are essential for the maintenance of pregnancy, fetal growth, and postpartum lactation. Some of the hormones produced during pregnancy are present in much larger amounts whereas others are in fewer amounts than in non-pregnant women. For example, the production of progesterone rises during pregnancy as needed not only for the establishment of pregnancy, but also for inhibition of ovulation and development of the breasts[1].

A study documented that expansion in plasma volume assists the transport of blood and nutrients to the fetus via the placenta[2]. Most of these changes begin soon after conception and continue until late gestation. An early report showed that these physiologic adaptations of pregnancy result in many significant changes in laboratory test values[2]. Some of these changes are well-known, such as the reduction in hematocrit and hemoglobin levels, which is termed physiologic or delusional anemia of pregnancy[3]. While the physiological increase in Glucose level occurs, is known as gestational diabetes of pregnancy[4].

This indicates that some of the blood parameters of pregnant women are found to be high/low, but they are not considered abnormal as they reverse after the termination of pregnancy. The blood parameters alter due to differences in race, nutrition, and gestational age, environmental and socio-economic factors[5]. Besides, no data is available for our healthy pregnant women, so the present study was designed to investigate some of the blood parameters during three trimesters of pregnancy in women of Abbottabad, Pakistan.

II. Materials and Methods:

A total of 90 pregnant women (60 primigravidas and 30 multigravidas) of middle socio-economic class were selected for the study. They were attending the OPD of the women and children hospital, Abbottabad for their routine antenatal visits. Twenty-five non-pregnant, non-lactating women at days 4-10 of normal menstrual cycle and belonging to the same socioeconomic background were also included as a control group.

All the participants were subjected to a questionnaire including age, height, weight, family members, occupation, history of ailments and medications, the onset of puberty, age at marriage, gestation period, and age at first delivery, the status of delivery, history of lactation, and parity. The study was approved by the local ethical research committee of the Women's Medical College, Abbottabad. The objectives of the project were disclosed to the participants who were assured of confidentiality.

The age of both pregnant and control volunteers was between 18 and 35 years. Those women who were residents of Abbottabad and willing to participate in the study were included in the study. All of them had regular ovulatory cycles, normal obstetric history, and never used any contraceptive during their reproductive life. While women with poor general health, having a smoking history or any chronic disease or any endocrinological disorder, and those who did not give birth to live healthy singleton infants at term were excluded.

Eight ml blood samples were collected in the morning between 08 and 09 a.m. during the first (1-13 weeks), second (14-26 weeks), and third trimesters (27+ weeks) of gestation. To 03 ml of blood EDTA was added and from the remaining blood after clotting serum was separated. Biochemical parameters were measured through automatic instrument Cobas 311 of Roche Company, Hematological by instrument CELL-DYN ruby Abbott Company while Architect plus I 1000 SR instrument of Abbott Company were used for estimation of hormones.

A statistical package for social sciences (SPSS) version 17 was used for the statistical analysis of the data. The level of significance was set at 0.05, and frequency distribution tables were created with means and standard deviations.

III. Results:

The hormonal parameters are presented in Table 1. The mean concentration of Progesterone was 42.1ng/ml during the first trimester, then rose to the level of 126.5ng/ml in the second trimester and again decreased in the third trimester (109.4ng/ml). The Estradiol levels increased steadily with the progress of the pregnancy.

Whereas the increased level of Prolactin hormone was observed in the third trimester and decreased level of Ferritin in the first trimester of pregnancy. Both Vitamin D3 and B12 were found lower in the pregnant women as compared to the control group of non-pregnant women.

Table -2 is about the biochemical changes during pregnancy. Glucose level was found high in the second trimester and then declined in the third trimester. Cholesterol and triglycerides were increased whereas urea, Creatinine, Protein, Albumin, Iron, and Calcium decreased during the three trimesters of pregnancy.

The hematological parameters of both the control and pregnant groups are shown in Table 3. The hemoglobin, red blood cells, Hematocrit, and Platelets were decreased as the pregnancy progressed. White blood cells ranged from 6.2 x 103 to 6.9 X 103 per cubic millimeter with a peak value in the mid-gestation. Moreover, a clear rise was observed in the percentage of Neutrophils through gestation.

Comparison of the blood parameters among pregnant and non-pregnant women are shown in Table-4. All the parameters were altered significantly with pregnancy except Calcium, creatinine, and platelets. Moreover, both the primigravidas and multigravidas of our study group had no more different pattern of all the investigated parameters. Besides, age at marriage and the other included variables had also no effect on the above estimated parameters in both the control and study groups.

Table-1: Hormonal parameters during Pregnancy

Analyte	Non-Pregnant Mean ± SD	1st trimester Mean ± SD	2nd trimester Mean ± SD	3rd trimester Mean ± SD
Progesterone (ng/ml)	01.5 ± 0.4	42.6 ± 13.8	126.5 ± 14.6	109.4 ± 14.1
Estradiol (pg/ml)	60.1 ± 7.8	1436.5 ± 108	4874.0 ± 278.1	5428 ± 436
Prolactin (ng/ml)	10.2 ± 2.5	142.5 ± 32.6	175.6 ± 56.4	323.8 ± 96.2
Ferritin (ng/ml)	130.1 ± 26.8	68.8 ± 11.6	116.4 ± 14.2	80.6 ± 9.4
Vitamin-D3 (ng/ml)	47.8 ± 12.2	31.5 ± 10.4	16.5 ± 6.8	14.2 ± 8.8
Vitamin B12 (pg/ml)	762.0 ± 79.4	336.2 ± 64.5	458.0 ± 98.2	362.4 ± 69.5

Table-2: Biochemical parameters during Pregnancy

Analyte	Non-Pregnant Mean ± SD	1st trimester Mean ± SD	2nd trimester Mean ± SD	3rd trimester Mean ± SD
Glucose (mg/dl)	91.2 ± 11.5	90.1 ± 12.4	108.2 ± 14.2	99.8 ± 22.1
Urea (mg/dl)	28.8 ± 2.2	26.6 ± 2.2	27.8 ± 2.2	27.4 ± 2.1
Creatinine (mg/dl)	0.7 ± 0.2	0.65 ± 0.1	0.7 ± 0.2	0.65 ± 0.2
Cholesterol (mg/dl)	182 ± 19.8	208 ± 21.2	239 ± 25.4	286 ± 36.5
Triglycerides (mg/dl)	155 ± 14.4	165 ± 16.8	228 ± 26.2	292 ± 32.4
Protein (g/dl)	7.6 ± 0.4	7.1 ± 0.3	6.7 ± 0.3	6.8 ± 0.4
Albumin (g/dl)	4.4 ± 0.2	4.3 ± 0.1	3.8 ± 0.1	3.7 ± 0.1
Iron (ug/dl)	41.2 ± 4.1	40.2 ± 3.5	38.6 ± 3.0	34.4 ± 4.1
Calcium (mg/dl)	8.7 ± 1.4	8.6 ± 2.0	8.2 ± 1.9	8.0 ± 1.8

Table-3: Hematological parameters during Pregnancy

Analyte	Non-Pregnant Mean ± SD	1st trimester Mean ± SD	2ndtrimester Mean ± SD	3rd trimester Mean ± SD
Hemoglobin (g/dl)	12.4 ± 1.1	11.8 ± 1.4	11.5 ± 0.9	11.6 ± 1.6
Leukocyte ($10^3/mm^3$)	6.4 ± 0.4	6.2 ± 0.5	6.9 ± 0.5	6.7 ± 0.3
Neutrophil (%)	50.4 ± 4.0	51.8 ± 4.4	56.5 ± 4.3	58.6 ± 5.4
Red blood cells ($10^6/mm^3$)	4.1 ± 0.4	3.9 ± 0.2	3.7 ± 0.2	3.8 ± 0.3
Hematocrit (%)	41.2 ± 0.5	37.2 ± 0.4	35.8 ± 0.5	39.5 ± 0.6
Platelets ($10^3/mm^3$)	2.79 ± 0.4	2.78 ± 0.6	2.76 ± 0.2	2.69 ± 0.5

Table-4: Comparison of blood parameters among non-pregnant and pregnant (Average of three trimesters) women

Analyte	Non-pregnant women Mean ± SD	Pregnant women Mean ± SD	p- value
Progesterone (ng/ml)	1.5 ± 0.4	92.8 ± 14.1	P< *0.0001*
Estradiol (pg/ml)	60.1 ± 7.8	3912.8 ± 274	P< *0.0001*
Prolactin (ng/ml)	10.2 ± 2.5	213.97 ± 61.7	p< *0.0001*
Ferritin (ng/ml)	130.1 ± 26.8	88.6 ± 11.7	p< *0.0001*
Vitamin-D3 (ng/ml)	47.8± 12.2	20.7 ± 8.7	p< *0.0001*
Vitamin B12 (pg/ml)	762.0 ± 79.4	385.5 ± 77.4	p< *0.0001*
Glucose (mg/dl)	91.2 ± 11.5	99.4 ± 16.2	p< *0.019*
Urea (mg/dl)	28.8 ± 2.2	27.3 ± 2.1	p< *0.002*
Creatinine (mg/dl)	0.7 ± 0.2	0.67 ± 0.2	P= *0.508*
Cholesterol (mg/dl)	182 ± 19.8	244.3 ± 27.7	p< *0.0001*
Triglycerides (mg/dl)	155 ± 14.4	228.3 ± 25.1	p< *0.0001*
Protein (g/dl)	7.6 ± 0.4	6.87 ± 0.3	p< *0.0001*
Albumin (g/dl)	4.4 ± 0.2	3.93 ± 0.1	p< *0.0001*
Iron (ug/dl)	41.2 ± 4.1	37.7 ± 3.5	p< *0.0001*
Calcium (mg/dl)	8.7 ± 1.4	8.3 ± 1.9	P=*0.329*

Hemoglobin (g/dl)	12.4 ± 1.1	11.63 ± 1.3	P=*0.0079*
Leukocyte (10^3/mm^3)	6.4 ± 0.4	6.6 ± 0.4	P=*0.029*
Neutrophil (%)	50.4 ± 4.0	55.63 ± 4.4	p< *0.0001*
Red blood cells(10^6/mm^3)	4.1 ± 0.4	3.8 ± 0.2	p< *0.0001*
Hematocrit (%)	41.2 ± 0.5	37.5 ± 0.5	p< *0.0001*
Platelets (10^3/mm^3)	2.79 ± 0.4	2.74 ± 0.4	P=*0.58*

IV. Discussion:

The measurements of different parameters in the maternal blood provide the right conditions for pregnancy success. In the present study, we observed a peak level of progesterone in the 2nd trimester which decreased again before term. Similar results were reported in an early study[6]. While some scientists observed a progressive increase in the concentration of progesterone during pregnancy[7].

The increase in the level of progesterone in the first trimester is derived largely from placental extraction of maternal cholesterol while in the third trimester, some progesterone is converted into 20 alpha-dihydro-progesterone, resulting decline in the concentration of the said hormone[6].

The fall in progesterone and the rise in estrogen in the third trimester in maternal plasma as observed in our study are considered prerequisites of parturition[8]. To explain the point that fall in progesterone level is necessary for delivery, some scientists, for premature delivery, treated lamps with 3 beta-hydroxysteroid dehydrogenase inhibitor injections which resulted in progesterone withdrawal, increased PGF2 alpha secretion and uterine contractions[9].

However, prolactin levels were progressively increased during gestation most probably due to its role in future lactation[10]. A study documented that estrogens have a stimulating effect on the secretion of prolactin[11]. Another study showed that prolong lactational amenorrhea in women is associated with higher prolactin levels[12].

Ferritin level progressively decreased from the first to the third trimester. This may be due to the increased need for iron which triggers ferritin mobilization from its stores. Similar results were shown in an early study[13]. This disagrees with the study done by others who showed that ferritin decreased from the first to the second trimester with a slight rise in the third trimester[14].

In the present study, it was found that pregnant women had deficiencies in vitamin D and Vitamin B12 levels in their blood. Similar results have been shown by the previous researchers[15]. The possible reason for the low level of vitamin D might be that the study populations have no more exposure to sunlight. Whereas Vitamin B12 in maternal blood might be low because of extreme fetal needs. A study documented that woman with a deficiency of vitamin B12 results in megaloblastic anemia while 25(OH) D levels below 15ng/ml had a fivefold increased risk of preeclampsia[15].

Glucose levels were found more in the second trimester of pregnancy. Similar results were reported earlier[16]. As pregnancy progresses, a well-integrated metabolic shift occurs for an adequate supply of nutrients to a constantly feeding fetus[16]. Besides, pregnancy is also associated with an insulin-resistant situation[16]. But some scientists claimed that glucose decreased in pregnancy due to the acute photolytic enzyme system of the placenta which increased the biosynthesis of insulin molecules[17].

Cholesterol and triglycerides in pregnant women were higher than in non-pregnant women. Similar results were shown in the previous study[16]. The increase in cholesterol and triglyceride levels observed in pregnant women may be because of changes in lipid metabolism during pregnancy due to an increase hormonal changes[6]. The increased level of progesterone as reported in our study affects metabolic changes which favor the biosynthesis of lipids[6]. Besides, serum triglyceride level increment in pregnancy may be due to the high concentration of many steroids that occur as normal pregnancy advances[16].

Protein, albumin, urea, and creatinine were found low as compared to non-pregnant women. Similar results were observed by others[2]. This might be because of increase plasma flow and glomerular filtration rate during pregnancy. The serum calcium and Iron decreased during the three trimesters of pregnancy like a previous study[17], most probably due to the dietary deficiency of the nutrients and increased demand of the mother and baby.

We found a progressive decline in hemoglobin, hematocrit, platelets, and red cells and a rise in leukocyte concentration from the first to the third trimesters of pregnancy. Similar findings were demonstrated by the previous scientists[18]. The decline may be due to an increased demand for iron as pregnancy progresses to fulfill fetal needs. A rising leukocytes count in pregnancy is not a reliable indicator of infection. Pain, nausea, vomiting, and anxiety have been reported to cause leukocytosis in the absence of infection[19].

Due to hemodilution secondary to the expansion of plasma volume, the platelets count in normal pregnancies might decrease[20]. The same study documented that after anemia, thrombocytopenia is the second most common hematological abnormality that occurs during pregnancy[20]. The interesting point observed in the present study is that women having low hemoglobin did not manifest clinical anemia nor did they complain of any anemic symptoms.

The findings of our study showed that there is a substantial difference in normal values of several blood parameters during pregnancy when compared with the non-pregnant women which is important information as needed for the interpretation of results. The limitation of this study was observed in budgeting. This study was not funded by any governmental, private, or non-profit funding bodies. The lack of budget restricted the research to limited samplings. The authors carried out this study for the best interest of the community and do not have any conflicts of interest to declare.

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