International Journal of Current Advanced Research
ISSN: O: 2319-6475, ISSN: P: 2319-6505, Impact Factor: SJIF: 5.995
Available Online at www.journalijcar.org
Volume 6; Issue 8; August 2017; Page No. 5225-5228
DOI: http://dx.doi.org/10.24327/ijcar.2017.5228.0676

Research Article

COMPARISON OF WBC- TOTAL AND DIFFERENTIAL COUNT IN DIFFERENT TRIMESTERS OF PREGNANCY

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ARTICLE INFO

Article History:

Received 16th May, 2017
Received in revised form 14th June, 2017 Accepted 24th July, 2017
Published online 28th August, 2017

Key words:

Pregnancy, Pregnancy trimester, WBC count

ABSTRACT

Background: - Pregnancy is a state characterized by changes in many physiological and haematological parameters. Most earlier studies conducted emphasized mainly on Haemoglobin and RBC parameters changes in pregnancy but there have been only a few studies on changes in WBC -total and differential count parameters in pregnancy. Leukocytosis in pregnancy is seen mainly due to neutrophilia with slight left shift along with toxic granulation associated with stress. The aim of this study was to assess the changes in total and differential leucocytes count in pregnancy in comparison with non-pregnant controls in different trimesters in pregnancy. The cross sectional study was conducted in Department of Pathology, Index Medical College Hospital and Research Centre, Indore during October 2013 to September 2015. A total 400 blood samples from pregnant women and non-pregnant women each were collected and run on fully automated 5part differential hematology analyser Transasia Model-XS-800i. Detail parameters including WBC- total and differential count were done and compared in different trimesters of pregnancy. In our study around two third females were in age group of 21-30 years. In total 39%, 38.75% and 22.25% women were in first, second and third trimester respectively. Comparison of mean between three trimesters was done. Total WBC count, Neutrophils and Lymphocytes count showed significant difference, while Monocytes, Eosinophils and Basophiles did not show any significant correlation. Significant increase in WBC and Neutrophil count were seen in pregnant women. There was significant difference in WBC count and neutrophils between first-second and first -third trimesters.

INTRODUCTION

Pregnancy is a state characterized by changes in many physiological and haematological parameters, which may be pathological in the non-pregnant women.[1] Many factors influence the pregnancy i.e. culture, environment, socioeconomic status and availability of medical care. These haematological parameters changes may also affect the pregnancy and its outcome.[2] Most of the studies conducted on haematological changes have given emphasis on haemoglobin concentration and RBC parameters changes in pregnancy, however only a handful of studies have been done on changes in WBC- total and differential count in pregnancy.[3,4] The white blood cells constitute about 1% of blood cells and are responsible for the immune system. There are five subsets of white blood cells, neutrophils, eosinophils, basophils, lymphocytes, and monocytes.[5] Increased WBC count is major finding seen in the pregnancy mainly associated with physiologic stress and increased inflammatory response associated with pregnancy.[6,7]

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This leucocytosis is mainly due to neutrophilia and slight neutrophilic left shift with toxic granulation. Monocytosis is also reported in pregnancy.[4] Several other changes also have been reported in neutrophils during pregnancy. They include, impairment of apoptosis due to the increased inflammatory response, reduced chemotaxis and impaired respiratory burst.[4, 7, 8, 9] Lymphocytes, eosinophils and basophils decline in number with increasing gestational age in pregnancy.[6, 10]

The aim of this study was to assess the changes in total and differential leucocytes count in pregnancy in comparison with non-pregnant controls in different trimesters in pregnancy.

MATERIAL AND METHODS

The cross sectional study was conducted in department of pathology, Index Medical College Hospital and Research Centre, Indore from October 2013 to September 2015. The total 400 blood samples from pregnant women and 400 blood samples from non-pregnant women were collected for the study. All the pregnant women of 20-40 years attending antenatal clinic with positive pregnancy test and live fetus in ultrasonography were included in the study. Pregnant women

sufferings from bleeding disorder, splenomegaly, hypertension, HIV, Hepatitis B and women on NSAID were excluded from study. After taking written consent, 2-3 ml blood from anticubital vein was collected by using dry, sterile disposable syringe and needle. Blood was immediately transferred in a tube containing anticoagulant ethylene-diamin-tetraacetic acid (EDTA). Detail history was taken. A drop of blood was placed on two slides and smears were prepared. The slides were stained with Leishman and field stain respectively. The collected samples were run in fully automated 5 part differential hematology analyser Transasia Model-XS-800i. Detail parameters including WBC- total and differential count were obtained and compared for different trimesters of pregnancy. Data was tabulated and analysed by using SPSS (v 16, IBM, Armonk, NY, USA). Mean, standard deviation, Pearson's Chi-square test and one way analysis of variance (ANOVA) were used for analytic assessment and differences were considered statistically significant when the P value was <0.005. The study was approved by the ethical review committee of the institute.

RESULTS

Table 1 summarizes the age classification which showed that 63 (19.75%) controls and 95 (23.75%) pregnant women were ≤20 years old. Age group 21-30 years comprised 266 (66.13%) controls, and 263 (65.75%) pregnant women. Controls and pregnant women aged >30 years old were 71 (14.13%) and 42 (10.50%). The difference between pregnant and non-pregnant women in term of age distribution was not significant (P=0.999). The mean ages of controls and pregnant women were 27.4±6.3 and 27.3±6.8 respectively. The independent sample t- test also showed no significant difference between mean ages of pregnant and non-pregnant women (P=0.991).

Table no 1 Distribution of patients according to age group

Age Group	Case Group		Control Group	
	No.	%	No.	%
<=20 years	95	23.75	63	19.75
21-30 years	263	65.75	266	66.13
31-40 years	42	10.50	71	14.13
Total	400	100.0	400	100.00

Comparison between Case Group and Control Groups

There was a significant increase in WBC count in pregnant women compared to non-pregnant women. Neutrophils are significantly increased in pregnant women compared to non-pregnant women. (Table No.2)

Table No. 2 Comparison of haematological value between case and Control Groups

Parameter	Case Group (n=400)	Control Group (n=400)	't' Value	P Value
	(Mean±SD)	(Mean±SD)		
WBC	9881±3759	8729±21.89	5.30df=798	0.000*
Neutrophil	73.06±7.98	64.86±6.10	16.33df=798	0.000*
Lymphocyte	21.62±7.07	27.54±6.19	-12.61df=798	0.000*
Monocytes	3.41±1.38	4.75±1.20	-14.73df=798	0.000*
Eosinophil	1.94±2.06	3.00±1.19	-8.96df=798	0.000*
Basophil	0.00±0.00	0.00±0.00	-	-

Comparison between Three Trimesters of Case Group

Out of 400 pregnant women 156 (39%), 155(38.75%) and 89 (22.25%) women were first, second and third trimester respectively. (table No.3) Comparison of mean between three trimesters done and Total WBC count, neutrophils and lymphocytes count showed significant difference, while monocytes, eosinophils and basophiles did not show any significant correlation.(Table No.4)

Table No 3 Distribution of case group patients according to trimester

Trimester	No.	%
First Trimester	156	39.00
Second Trimester	155	38.75
Third trimester	89	22.25
Total	400	100.00

Table 4 Comparison of Mean WBC and Differential count between the groups

Parameter	First trimester (n=156)	Second trimester (n=155)	Third trimester(n=89)
	(Mean±SD)	(Mean±SD)	(Mean±SD)
WBC	9413±2314	10277±5137	10013±2790
Neutrophils	70.79±7.18	75.69±7.22	72.44±9.22
Lymphocytes	23.68±6.35	19.47±6.81	21.75±7.66
Monocytes	3.56±1.29	3.20±1.28	3.49±1.65
Eosinophils	1.90±1.30	1.77±1.82	2.29±3.21
Basophils	0.00±0.00	0.00±0.00	0.00±0.00

Comparison of Mean WBC between the groups

One –Way ANOVA test was used with F value=2.14, P value=0.119, Not significant. The F value obtained was 2.14 with a P value of > 0.05, which is statistically not significant. Thus, WBC values in all the three groups are comparable. As the ANOVA value was found to be non-significant, post-hoc Tukey test has not been applied. (Table No. 4)

Comparison of Mean Neutrophil count between the groups

One- Way ANOVA test was used with F value = 16.09, P value = 0.000, Significant.

The F value obtained was 16.09 with a P value of < 0.05, which is statistically significant.

Thus, neutrophil values in all the three groups are statistically different. (Table No.4)

Significant difference was seen between the pairs first trimester- second trimester, and second trimester- third trimester, while non-significant difference was seen between first trimester-third trimester pair. Post- hoc Tukey was applied to see the difference between the pairs. (Table No. 5)

Table No. 5 Post-hoc Tukey Test was applied to see the difference between the pairs

Pair	Mean Difference	't' Value	P Value
First trimester- Second trimester	4.89	5.61	0.000*
First trimester- Third trimester	1.65	1.62	0.238
Second trimester- Third trimester	3.24	3.17	0.004*

*- Significant difference

Comparison of Mean Lymphocyte count between the groups

One-Way ANOVA test was used, F value = 14.79, P value = 0.000, Significant

The F value obtained was 14.79 with a P value of < 0.05, which is statistically significant.

Thus, lymphocyte values in all the three groups are statistically different. (Table No. 4)

Significant difference was seen between the pairs first trimester- second trimester and second trimester-third trimester, while non- significant difference was seen between first trimester-third trimester pair. Post-huc Tukey Test was applied to see the difference between the pairs. (Table No. 6)

Table No. 6 Post-huc Tuskey Test was applied to see the difference between the pairs

Pair	Mean Difference	't' Value	P Value
First trimester- Second trimester	4.21	5.44	0.000*
First trimester- Third trimester	1.93	2.12	0.086
Second trimester- Third Trimester	2.29	2.52	0.032*

*- Significant Difference

Comparison of Mean Monocyte count between the groups

One-Way ANOVA test was used. F value = 2.96, P value = 0.053, Not Significant

The F value obtained was 2.96 with a P value of > 0.05, which is statistically not significant.

Thus, monocytes values in all the three groups were comparable. As the ANOVA value was found to be non-significant, Post-hoc Tukey test has not been applied. (Table No. 4)

Comparison of Mean Eosinophils between the groups

One-Way ANOVA test was used. F value = 1.87, P value = 0.155, Not Significant.

The F value obtained was 1.87 with a P value of > 0.05, which is statistically not significant.

Thus, eosinophils in all the three groups are comparable. As the ANOVA value was found to be non-significant, Post-hoc Tukey test has not been applied. (Table No. 4)

Comparison of Mean Basophils between the groups

For Basophils One- Way ANOVA test could not be applied.

DISCUSSION

Our study shows increases in total WBC count in pregnant women as compared to the control group and this is similar to the finding of study conducted by the Kuhnert *et al,* [11] Osunga *et al,* [12] Luppi *et al,* [13] Osoagabaka *et al,* [14] James *et al,* [15] Cunningham *et al,* [16] Chandra *et al,* [6] Das *et al,* [17] and Elgari *et al.* [18] The increase in total WBC in pregnant women is result of body building the immunity of the fetus and it is achieved by a state of selective immune tolerance, immunosuppression, and immunomodulation in the presence of a strong antimicrobial immunity. There is also down regulation of potentially dangerous T-cell-mediated immune responses, while activating certain components of the innate immune system, such as neutrophils. This unique dysregulation between different components of the immune system plays a central role in the maternal adaptation to pregnancy. However Ichipi-Ifukor *et al* did not report significant changes in total WBC count in pregnant in comparison of non-pregnant control group women.[19]

Our study does not show relation between increased total WBC count and increase in gestation age. This is probably because our study is cross sectional study with varying number of subjects at different gestational age. These finding are similar to the Pughikumo *et al,* [20] Osonuga *et al.*[12] Increased TWBC with increased gestation age was however seen by pervious workers like Crocker *et al,* [8] Osoagabaka OU *et al,* [14] and Lurie S *et al.* [21]

Our study shows neutrophilia which is similar to the study by Fleming AF *et al* [22] and Gatti L *et al* [23]. This is due to impaired neutrophilic apoptosis during pregnancy [23]. The neutrophils shows toxic granulation and left shift with increase in immature cells like myelocytes and metameyelocytes in the peripheral smears.[24] The Neutrophil count rises with gestation age and may remain elevated throughout the pregnancy. Significant difference was seen between pairs first trimester-second trimester and second trimester-third trimester, while non-significant difference was seen between first trimester-third trimester pair.[25] Our study show increased neutrophil count with gestation age.

The lymphocyte count decreases with increased gestational age as reported by the various studies is also comparable with our study. Few studies also reported increase in lymphocyte count in pregnant women.[12,26] However no significant difference was observed in different trimesters.[12] Purohit *et al* reported that lymphocytes count decreases significantly during first and second trimesters and but increase during the third trimester. [25]

There is an absolute monocytosis during pregnancy especially in the first trimester but it decreases with advance in gestation. Monocytes help in preventing foetal allograft by infiltrating the decidual tissue (7th-20th week of gestation) possibly, through PGE2 mediated immunosuppression. [13, 27] Study by Pitkin RM *et al* also show Monocytosis but our study does not shows any significant changes in monocyte count.[10]

Basophils and eosinophils decreases in pregnant women in comparison with control cases.[3, 16] Our study does not shows any significant changes in Basophils and Eosinophils similar to the study of Edlestam G *et al.* [28] However Das S *et al* reported increased in Eosinophilic count in their study.[17]

We can conclude that our study showed increase in total WBC and neutrophil count in pregnant women. Neutrophils and lymphocytes count shows significant changes in between first-second and second-third trimesters.

CONCLUSION

There was a significant increase of total WBC count and Neutrophil in pregnant women than non-pregnant women. There was significant difference in neutrophils and lymphocytes between first-second and second-third trimesters. Monocytes, Eosinophil and Basophils do not show any significant changes in pregnant women.

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How to cite this article:

Somendra Kumar Dhariwal *et al* (2017) 'Comparison of Wbc- Total and Differential Count in Different Trimesters of Pregnancy ', *International Journal of Current Advanced Research,* 06(08), pp. 5225-5228.
DOI: http://dx.doi.org/10.24327/ijcar.2017.5228.0676

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