Revista Argentina de Clínica Psicológica
2020, Vol. XXIX, N°4, 282-286
DOI: 10.24205/03276716.2020.827

Establishment of Reference Intervals for Complete Blood Count Parameters During Normal Pregnancy in Second Trimester

Zhang Min*[a], Lu Hui-Xia[a], Zhang Zheng-Kang[a], Zhang Qiang[a]

Abstract

Background: Pregnant women belong to the normal population, they have their unique features, including undergoing a series of bodily changes during the pregnancy. However, there are no reference intervals of pregnant women in China. In this study, reference intervals for complete blood count parameters of Chinese pregnant women was established.

Methods: Healthy pregnant women took the blood tests at second trimesters. Healthy adult women took the tests at the same time. All blood samples were tested on Sysmex XE-2100.The following CBC parameters were analyzed: red blood cell count (RBC), hemoglobin (HB), hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), platelet count (PLT), white blood cell count (WBC), and leukocyte differential classification. Determine whether there is a difference between the pregnant women and healthy adult women. Reference intervals were calculated based on the $\bar{x} \pm 1.96s$.

Results: The RBC count, HB concentration, HCT and PLT of pregnant women were significantly lower than values for healthy adult women ($p<0.001$). MCV, MCH, MCHC,and WBC count in pregnant women were significantly higher than in healthy adult women ($p<0.001$). The percentage of NEUT increased significantly; the percentage of MONO increased slightly; and the percentages of LYMPH, EO, and BASO all decreased accordingly. The reference intervals of which for RBC was 3.19-4.58×10^{12}/L , Hb was 100.04-139.43 g/L, HCT was 0.32-0.38 and for PLT was 91.27-281.32×10^{9}/L. The reference intervals of which for MCV was 81.40-99.83fL, MCH was 24.95-36.85pg, MCHC was 320.90-358.90g/L. The reference intervals of which for WBC was 5.20-13.59×10^{9}/L.NEUT% was 63.34%-84.51%, LYMPH% was 10.21%-29.18%, MONO% was 2.96%-7.91%, EO% was 0%-2.36%, BASO %was 0%-0.46%.

Conclusions: There are significant difference between the pregnant women and healthy adult women for CBC. The changes of CBC parameters during pregnancy are described. The reference intervals for pregnant women in second trimester are determined in this study.

Keywords: complete blood count; pregnancy; reference interval.

1. Introduction

Pregnant women are part of the normal The reference intervals of which for RBC was 3.19-4.58×10^{12}/L , Hb was 100.04-139.43 g/L, HCT was 0.32-0.38 and for PLT was 91.27-281.32×10^{9}/L. The reference intervals of which for MCV was 81.40-99.83fL, MCH was 24.95-36.85pg, MCHC was 320.90-358.90g/L. The reference intervals of which for WBC was 5.20-13.59×109/L.NEUT% was 63.34%-84.51%, LYMPH% was unique features, including undergoing a series of bodily changes during the pregnancy. For example, the total blood volume of a pregnant women is 30% greater than that of a non-pregnant woman.

The increase in the plasma volume of pregnant women occur more rapidly than the proliferation of

a*. Department of Blood Transfusion, Zhongda Hospital, School of Medicine, Southeast University. Nanjing. People's Republic of China
***Corresponding Author: Zhang Min**
Email: zhangmi0076@sina.com

red blood cells (RBC), which leads to blood dilution and causes physiologic anemia [1–3]. In addition, the number and concentration of white blood cells (WBC), platelets（PLT）, and coagulation factors in pregnant women change remarkably. As a result, the blood indices of pregnant women are significantly different from that of non-pregnant adult women.

Considering that reference intervals are designed to assist clinicians during diagnostic triage, criteria must reflect the nature of the subjects entering the system for care. The reference intervals of complete blood count (CBC) were significantly different due to the different characteristics of the population. For example, men and women possess different reference intervals of CBC;the hemoglobin (HB) concentration and RBC count exhibit significant differences during the various stages of newborn, juvenile and adolescent; the ratio of granulocytes and lymphocytes also exhibits regular changes; and pregnant and non-pregnant women have different reference intervals of CBC. It is necessary to establish a reference interval according to the physiologic characteristics of different populations [4]. We should establish appropriate reference interval of CBC for pregnant women.This help us distinguish the physiologic and pathologic changes of pregnant women effectively[5].

The World Health Organization (WHO) has clarified the blood index boundary value for pregnant women, non-pregnant women, and children under 5 years of age in "The management of nutrition in major emergencies" [6]. Some related studies have also reported reference interval s of CBC for pregnant women [7,8]. However, these indices are affected by ethnics, geographic, social and economic environments and social status [9], limiting their application in China. The trend of medical development is precision and individualization.It is necessary to establish a standard reference interval of CBC for pregnant women. In this study, reference interval was established based on the number of CBC in healthy pregnant women. These data provide the basis for the diagnosis and treatment of pregnancy health care and diseases.

2. Materials and methods

Pregnant women who registered in our hospital obstetric clinic and received perinatal health-care from August to October 2016 were selected as research subjects. The 700 cases selected were aged between 19 and 45 years, were at 20–24 weeks of gestation.They were all healthy and without any underlying disease history (heart, lung, liver, kidney, or blood disease). They have no abnormal symptoms such as infection and fever at the time of blood collection. At the same time, 700 healthy adult women in the hospital were selected for physical examination as controls.

Each venous blood sample was collected into an EDTA-containing vacuum blood collection tube (Becton Dickinson Medical Devices Co Ltd, NC, USA). Professional technical personnel in our laboratory department analyzed the specimens within 1 hour. Data was processed by Sysmex XE-2100(Sysmex Corporation, Kobe, Japan). Three levels of internal quality control（IQC）were made every day. We participate in external quality assessment (QEA) regularly. The results are qualified.

SPSS 20.0 software (SPSS Inc., Chicago, IL, USA) was used to perform statistical analyses. Quantitative data were presented as $\bar{x}\pm s$. A t test was applied when the data showed a normal distribution and homogeneity of variance. A corrected t test was used when the data were normally distributed and showed homoscedasticity. The significance level was $\alpha=0.05$. Differences were significant when $p<0.05$. Index reference intervals were calculated based on the $\bar{x}\pm 1.96s$.

3. Results

The RBC count, HB concentration and hematocrit (HCT) of pregnant women were significantly lower than values for healthy adult women ($p<0.001$). Mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) in pregnant women were significantly higher than in healthy adult women ($p<0.001$). Data are shown in Table 1.

The WBC count in pregnant women was substantially higher than in healthy adult women ($p<0.001$). The percentage of neutrophils (NEUT) increased significantly; the percentage of monocytes (MONO) increased slightly; and the percentages of lymphocytes (LYMPH), eosinophils (EO), and basophils (BASO) all decreased accordingly. PLT was significantly lower than value for healthy adult women ($p<0.001$). Data are shown in Table 2.

4. Discussion

The existence and development of appropriate laboratory medicine therefore aim at accuracy and personalization, such that clinicians may be provided with accurate data for proper diagnosis. The importance of reference interval is the same as the importance of laboratory test. Test is meaningless without it. When the reference interval is too large, it can result in false negative and some diseases may go

undetected. When it is too small, false positive may result, and excessive treatments may be performed.

Table 1. Anemia indices for pregnant and healthy adult women.

Index	Control group	Experimental group	t	p
RBC(×10^{12}/L)	4.48±0.31	3.89±0.36	33.2	<0.001
HB(g/L)	131.11±11.36	119.73±10.05	19.85	<0.001
MCV(fL)	87.97±5.03	90.62±4.70	10.17	<0.001
MCH(pg)	29.29±2.18	30.90±3.03	11.41	<0.001
MCHC(g/L)	332.66±10.38	339.90±9.69	13.48	<0.001
HCT(L/L)	0.39±0.03	0.35±0.03	25.11	<0.001

Table 2. Infection indices for pregnant and healthy adult women.

Index	Control group	Experimental group	t	p
WBC(×10^{9}/L)	6.53±1.72	9.40±2.14	27.67	<0.001
PLT(×10^{9}/L)	240.44±53.23	186.29±48.48	19.9	<0.001
NEUT(%)	57.61±8.27	73.92±5.41	43.66	<0.001
LYMPH(%)	36.05±7.71	19.70±4.84	44.64	<0.001
MONO(%)	5.12±1.34	5.27±1.35	2.05	0.041
EO(%)	2.00±1.61	0.96±0.72	15.64	<0.001
BASO(%)	0.22±0.26	0.18±0.14	4.11	<0.001

The reference intervals of normal distribution data were calculated according to $\bar{x}\pm$ 1.96s. There were difference in reference intervals between the experimental group and the control group. The reference intervals are shown in Tables 3 and 4.

Table 3. Reference range for women with pregnancy anemia.

Index	Reference interval	
	Control Group	Experimental Group
RBC(×10^{12}/L)	3.87-5.10	3.19-4.58
HB(g/L)	108.84-153.37	100.04-139.43
MCV(fL)	78.11-97.83	81.40-99.83
MCH(pg)	25.05-33.55	24.95-36.85
MCHC(g/L)	312.32-353.00	320.90-358.90
HCT(L/L)	0.36-0.42	0.32-0.38

Table 4. Reference range for infected pregnant women.

Index	Reference interval	
	Control Group	Experimental Group
WBC(×10^{9}/L)	3.16-9.90	5.20-13.59
PLT(×10^{9}/L)	136.11-344.78	91.27-281.32
NEUT(%)	41.41-73.82	63.34-84.51
LYMPH(%)	19.94-50.16	10.21-29.18
MONO(%)	2.79-7.75	2.96-7.91
EO(%)	0-5.16	0-2.36
BASO(%)	0-1.58	0-0.46

Reference interval that is neither too large nor too small thus benefits treatment strategies. At present, there are no reference intervals of CBC for pregnant women in China. Most doctors take the reference intervals of normal adult women as a reference and combine clinical experience to determine whether there are any diseases in pregnant women. Anemia is the most common complication observed during pregnancies, increasing the risk for women and their neonates. Anemia is also associated with placental oxygen deficiency, limited growth, low-weight infants, premature birth, stillbirth, and intrauterine fetal distress [10]. The diagnostic standard for pregnancy anemia was first established by the nutritional anemia study group of the WHO in 1968 [11–13]. Internationally, researchers recommended the WHO standard in 1972, which denoted pregnancy anemia as a HB concentration less than 110 g/L [14]; and the Center for Disease Control standard depicted pregnancy anemia as a HB concentration lower than 110 g/L during early pregnancy, 105 g/L during mid-pregnancy, or 110 g/L during late pregnancy in 1998[15]. It was not clear as to whether this large data from 18 countries set fit all individuals living in the different countries, which encompassed different ethnic groups and living conditions and habits. Sarah K[16] explicitly indicated that pregnant women of different ethnicities showed significantly different blood indices. China has a vast area, there are 56 ethnic groups. Different ethnic groups are distributed in different regions. It is significant to establish regional reference interval of

CBC for pregnant women in china.Our data showed that because of blood dilution, the RBC count and HB concentration, and HCT of pregnant women were significantly lower than in healthy adult women. In addition, mean value of MCV, MCH, and MCHC were significantly augmented in pregnant women.However, blood dilution cannot explain the phenomenon in pregnant women. Research shows 44% of women had plasma folate deficiency, and 35% had red cell folate deficiency[17]. At the beginning of 2002, 10 of the 17 countries had an official government recommendation that women planning a pregnancy should take folic-acid supplementation daily[18].As can be seen, folate deficiency is common in pregnant women.The increase in MCV, MCH, and MCHC might be caused by folate deficiency. According to the reference-established rule of $\bar{x} \pm 1.96S$, our study established reference intervals for RBC, HB, MCV, MCH, and MCHC in pregnant women; the reference intervals of which for RBC was $3.19\text{-}4.58\times10^{12}$, HB was 100.04–139.43 g/L, and for HCT was 0.32-0.38. These indices in Chinese pregnant women can be lower than healthy adult women. Standard of anemia recommended by WHO is not suitable for pregnant women in China. Therefore, the diagnostic standard for anemia should be RBC count below 3.19×10^{12}, HB below 100.04 g/L, or HCT below 0.32. To prevent excessive iron supplementation in normal pregnant women, increasing physical and economic burden.

The reproductive and endocrine systems, and metabolism of pregnant women undergo a count of changes during pregnancy. WBCs start to proliferate from 7–8 weeks and reach a maximum at 30 weeks, with the majority of WBCs being neutrophils. This is a complicated physiologic phenomenon, and its mechanism(s) remains unclear [19]. Impaired neutrophil apoptosis may explain the neutrophilia associated with normal pregnancy[20]. Pain, nausea, vomiting, and anxiety, which one woman would experience all of them during pregnancy, have been reported to cause leukocytosis in the absence of infection[21]. The change in the WBC population is closely associated with different organs, and this cell population is the most sensitive to various stimuli. The increase in the WBC count during pregnancy might be related to the release of WBC from marginal pools. Infection is one of the most common complications during pregnancy, and the WBC count is a critical index used to identify infections and to evaluate the severity of the infection. Thus, it is important to identify whether the increase in WBCs is due to a pathologic or physiologic reason, to evaluate whether the pregnant woman has an infection, and to assess the severity of the infection. In the present study, pregnant women exhibited a significantly higher level of WBC compared to healthy adult women ($p<0.001$), with a reference interval of $5.20\text{-}13.59\times10^{9}$. The most elevated population within the WBC was neutrophils, with a significant increase in the ratio; while the ratio of other types of WBC decreased. Furthermore, based on NEUT and LYMPH percentages (63.34%–84.51% and 10.21%–29.18%, respectively), we can determine whether the infection is of bacterial or viral origin. The granules in the neutrophils can then also be further used to evaluate the severity of the infection [22]. In addition, our study showed that PLT count was reduced during pregnancy, possibly due to the increase in total blood volume during pregnancy and concomitant blood dilution; as well as PLT collection, utilization, and increasing damage during placental cycles. Other possible mechanisms include the inhibitory effect of hormones on macrophages, and the fact that elevated estrogen levels during pregnancy enhance phagocytosis and the destructive effect of liver cells on platelets.

In conclusion, we determined reference intervals for CBC on pregnant women. The establishment of this reference intervals provide a powerful diagnostic and treatment tool for use by clinicians.

References

[1] Gou WL, Song Q. Interaction of pregnancy and anemia. Journal of Practical Obstetrics and Gynecology, 2003,19(5):129-130.

[2] Cao ZY, Lang JH, Dong Y, et al. Chinese obstetrics and gynecology[M]. Beijing: People Medical Publishing House, 1999.

[3] Milman N, Bergholt T, Byg KE, et a1. Reference intervals for haematological variables during normal pregnancy and postpartum in 434 healthy Danish women[J]. Eur J Haematol, 2007, 79(1): 39-46.

[4] Ritchie RF, Palomaki G. Selecting clinically relevant populations for reference intervals. Clin Chem Lab Med. 2004, 42(7): 702-709.

[5] Klajnbard A, Szecsi PB, Colov NP, et al. Laboratory reference intervals during pregnancy, delivery and the early postpartum period. Clin Chem Lab Med. 2010, 48(2):237-248.

[6] The management of nutrition in major emergencies. Geneva, World Health Organization, 2000. (http://whqlibdoc.who.int/publications/2000/9241545208.pdf, consultado el 7 Junio 2011).

[7] CDC. Recommendation to Prevent and Control Iron Deficiency in the United States. Morbidity

and mortality weekly report. 1998, 47(RR-3):1-12.

[8] WHO, UNICEF, UNU. Iron deficiency anaemia: assessment, prevention and control, a guide for programme managers. Geneva, World Health Organization, 2001.http://www.who.int/nutrition/publications/micronutrients/anaemia iron deficiency/WHO NHD 01.3/en/index.html,consultado el 7 Junio 2011).

[9] Harm SK, Yazer MH, Waters JH. Changes in hematologic indices in caucasian and non-caucasian pregnant women in the United States . Korean J Hematol.2012.47(2):136-141.

[10] Xie Xing, Gou Wenli. Obstetrics and Gynecology. Beijing: People Medical Publishing House, 2013. 92-93.

[11] Nutritional anaemias. Report of a WHO Scientific Group. Geneva, World Health Organization, 1968.(WHO Technical Report Series, No. 405).(http://whqlibdoc.who.int/trs/WHO TRS 405.pdf, consultado el 7 Junio 2011).

[12] World Health Organization. Worldwide prevalence of anemia 1993– 2005 [EB/OL]. Geneva: World Health Organization, 2008[2014-04- 12]. http://whqlibdoc.who.int/publications/2008/9789241596657_ eng.pdf.

[13] World Health Organization. Hemoglobin concentrations for the diagnosis of anemia and assessment of severity [EB/OL]. Geneva: World Health Organization, 2011[2014-04-12]. http://www.who.int/vmnis/indicators/haemoglobin.

[14] World Health Organization. Nutritional anaemias. Report of a WHO group of experts. World Health Organ TechRep Ser, 1972, 503: 1-29.

[15] Centers for Disease Control and Prevention. Recommendations to prevent and control iron deficiency in the UnitedStates[J]. MMWR Recomm Rep, 1998, 47(RR-3): 1-29.

[16] Harm SK, Yazer MH, Waters JH. Changes in hematologic indices in caucasian and non-caucasian pregnant women in the United States. Korean J Hematol, 2012, 47(2):136-141.

[17] Ren A, Zhang L, Li Z, et al. Awareness and use of folic acid, and blood folate concentrations among pregnant women in northern China--an area with a high prevalence of neural tube defects. Reprod Toxicol. 2006, 22(3): 431-436.

[18] Meijer WM, de Walle HE. Differences in folic-acid policy and the prevalence of neural-tube defects in Europe; recommendations for food fortification in a EUROCAT report. Ned Tijdschr Geneeskd. 2005, 149(46): 2561-2564.

[19] Odhiambo C, Omolo P, Oyaro B. Establishment of reference intervals during normal pregnancy through six months postpartum in western Kenya. PLoS One. 2017 Apr 11;12(4):e0175546. doi: 10.1371/journal.pone.0175546. eCollection 2017.

[20] Von Dadelszen P, Watson RW, Noorwali F, et al. Maternal neutrophil apoptosis in normal pregnancy, preeclampsia, and normotensive intrauterine growth restriction. Am J Obstet Gynecol. 1999,181(2):408–414.

[21] Milhorat AT, Small SM, Diethelm O. Leukocytosis during various emotional states. Arch Neurol Psychiatry. 1942,47(5):779–792.

[22] Edelstam G, Löwbeer C, Kral G, et al. New reference values for routine blood samples and human neutrophilic lipocalin during third-trimester pregnancy. Scand J Clin Lab Invest. 2001,61(8):583-592.