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MEASUREMENT OF PLASMA HOMOCYSTEINE LEVEL AMONG SUDANESE PREGNANT LADIES

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ABSTRACT
Background: There is many changes during pregnancy from first to third trimester if not following carefully may have an adverse effect both mother and baby life, one of these varibles is homocysteine amino acid (Hcy) this amino acid is methylated by MTHFR enzyme and B12 and folate as co-factor to give methionine which is important in DNA formation. During pregnancy methionine may be actively transported to the developing embryo that may decrease homocysteine in maternal plasma. **Objectives:** The aim of this study is to measure the plasma homocysteine level among sudanese pregnant ladies and to correlate homocysteine level with lady clinical data and age. **Material and methods**: A cross sectional study was performed in Khartoum state, sudan. A total of 30 EDTA blood sample collected from normal pregnant ladies in first trimester were included in this study**,** their age is ranged between 18 and 38 years, plasma homocysteine level was masuread by immune assay ELISA. The data were compared by using statistical analysis performed with statistical package for social sciences (SPSS) version 11.5 by using one sample T test. **Result:** The study showed that the mean of plasma homocysteine level in pregnant ladies at first trimester by micromoles per liter is (4.093) which was significantly lowered compared to normal range 5-14µmol/L (*P*-value0.00). **Conclusion:** Plasma homocysteine level is decreased during first trimester of gestation.

KEYWARDS: Hcy, pregnancy, plasma homocysteine, first trimester, MTHFR, B12, Folate.

INTRODUCTION
Pregnancy is the term used to describe the period in which a woman carries a fetus inside of her in most cases the fetus grows in the uterus.

Pregnancy usually lasts about 40 weeks, or just over 9 months, as measured form the last menstrual period to childbirth. pregnancy is divided in to three trimester, the major events in each trimester are described below.[1]

Frist trimester(week 1 to week 12)
The events that lead to pregnancy being with conception, in which the sperm penetrates the egg produced by ovary. the zygote is (fertilized egg) then travels through the woman,s fallopian tube to uterus, where it implants itself in the uterine wall. the zygote is made up of a cluster of cells formed from the egg and sperm. these cells from the fetus and placenta. the placenta provides nutrients and oxygen to the fetus.[2]

Second trimester (week 13 to week 28)
At 16 week and some time as early as 12 weeks, a woman can typically find out the sex of her infant, muscle tissue, bone and skin have formed.

At 20 weeks, footprints and fingerprints have formed and the fetus sleeps and wakes regularly.

Some researches supported that babeis born at 28 weeks was 92% although those born at this time will likely still experience serious health complications, including respiratory and heart problems.[3]

Third trimester (week 29 to week 40)
At 32 weeks, the bones are soft and yet almost fully formed and the eyes can open and close. Infants born before 37 weeks are considered pre-term, these children are at increased risk for problems such as developmental delays vision and hearing problems and cerebral palsy.[4]

Infants born in the 37th and 38th weeks of pregnancy - previously- considered full term, but now considered early term, this infants face more health risks than infants who are born at 39 weeks or later, which is now considered full term.[5]

Infants born at 39 and 40 weeks considered as full term. Infants born at 41 weeks through 41 weeks and 6 days are considered late term. Infants born at 42 weeks and beyond are considered post term.[4][5]

Homocysteine

Homocysteine (Hcy) is a four-carbon amino acid with a free thiol group, which is formed by demethylation of methionine, an essential amino acid derived from diet. Normal total Hcy (tHcy) concentration range from 5-14 µmol/L. Hyperhomocysteinemia (HHcy) has been classified into moderate (plasma tHcy concentrations of 15-30 µmol/L), intermediate (plasma tHcy concentrations of 31-100 µmol/L) and severe (plasma tHcy concentrations 100 µmol/L). Both acquired and genetic factors can have an impact on plasma tHcy. Male gender, aging, smoking, impaired renal function, and some medications such as Corticosteroids and Cyclosporine are some examples of the acquired causes and classic homocytinuria and C677T homozygote mutation of 5,10-methylenetetrahydrofolate reductase (MTHFR) are the main genetic ones. Vitamin B12, vitamin B6 and folate, all of which have dietary origins, are three main cofactors in Hcy metabolism. Deficiencies in these supplements are more prevalent in the most developing countries and may account for many cases of moderate hyperhomocysteinemia.[6]

Plasma homocysteine is normally lower in all 3 trimesters of pregnancy than in the non-pregnant state. Homocysteine concentration can be directly correlated with albumin concentration, which decrease during pregnancy and decrease further in pregnant women taking folic acid supplements. Because concentration of methionine were higher and those of homocysteine were lower in amniotic fluid than in maternal serum, these investigators suggested that methionine may be actively transported to the developing embryo.[7]

MATERIAL AND METHODS

A cross sectional study carried out at faculty of Medical Laboratory science, Al-Neelain Universtiy with sample comprised of 30 sudanese normal pregnant ladies at first trimester in Khartoum state, sudan. Pregnant ladies at second, third trimester and pregnant ladies with diseases and complication that affected the plasma homocysteine level were excluded.

Blood specimens were taken from normal pregnant ladies in first trimester, 3 ml of venous blood was collected in EDTA container, then blood samples were centrifuged and the plasma transferred to new plain containers.

immune assay ELISA was used to measure plasma homocysteine level :standard dilution was prepared then prepar the sample dilution and pipatting 40µL in testing well then covered with adhesive strip and incubated for 30 min at $37c^0$, centrifuged, washed and added enzyme (HRP conjugate reagent) except blank well, then incubated, washed and added 50µL chromogen solution B to each well after that added 50µL of stop solution to each well, finally the absorbance read at 450 nm within 15 min after adding the stop solution.

RESULT

The results of the present study showed that the mean of plasma homocysteine of pregnant ladies during first trimester by micromoles per liter was (4.093) which was low according to normal range (5-14µmol/L) as shown in Table 1.

Also the results showed their significantly lowered compared to non-pregnant ladies (*P*- value 0.000) as shown in Table 2.

The result of correlation of plasma homocysteine and the age of the study population showed no statistically significant correlation (*P*-value 0.590) as shown in Table 3.

Table 1: One-Sample Statistics of homocysteine and study group.

	N	Mean	Std. Deviation	Std. Error Mean
Homocysteine µmol/L	30	4.093	1.5243	.2783

Table 2: One-Sample Test.

	Test Value = 10					
	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
Homocysteine µmol/L	-21.225	29	.000	-5.907	-6.476	-5.337

Table 3: Correlation of plasma homocysteine and age.

		AGE	Homocysteine µmol/L
AGE	Pearson Correlation	1	0.102
	Sig. (2-tailed)	.	0.590
	N	30	30
Homocysteine µmol/L	Pearson Correlation	0.102	1
	Sig. (2-tailed)	0.590	.
	N	30	30

DISSCUSION

This study was carried out in Khartoum, Sudan. Aimed to measure the plasma homocysteine level among Sudanese pregnant ladies, its included 30 normal pregnant ladies at first trimester.

The present study revealed that decrease plasma homocysteine level (4.093±1.52) with confidance interval 95%, which was statistically significant compared to normal range(5-14µmol/L) (p value 0.000). this study was agree with three other studies first study is: Steegers-Theunissen *et al* in 1997 who reported that alterations in the methionine requirement might explain the reduction in tHcy concentrations in pregnancy in addition to hormonal influences and heamodilution. It is difficult, however, to see how increased utilization of methionine by the fetus at 8 wk of pregnancy would decrease maternal tHcy concentrations in early pregnancy.[8] Second study by Mark.C *et al* in 1999 who measured plasma homocysteine in 155 normal pregnant women and they found that plasma homocysteine is decreased during pregnancy (mean in micromoles per liter was 5.6 at 8-16 weeks of gestation, p-value 0.001).[9] Third study is Cikot *et al* in 2001 who reported only a slight reduction in tHcy concentration during early pregnancy, with no further decrease throughout mid-to-late pregnancy.[10]

The analysis result of this study showed that no statistically significant correlation between the age of pregnant ladies and plasma homocysteine level (p-value 0.590).

The Major limitations in the present study are the small sample size, and relatively short study period.

CONCLUSION

In summary we conclude that pregnant ladies at first trimester have decrease plasma homocysteine level and this change is not influanced by the age.

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