Ibrahim Card Med J 2017; 7 (1&2): 84-91

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Association between Food Behaviour of Pregnant Women and Status of Serum Vitamin B_{12} and Neonatal Outcome – A Cross-Sectional Study

Rowshan Afrooz [1], Lipika Ghosh,[2] Abul Kalam Md. Faruq,[3] Mitheel-Ibna Islam [4]

ABSTRACT:

Background & objective: To find the association between serum vitamin B_{12} and food behavior of the pregnant women and its influence on perinatal outcome.

Methods: This cross-sectional study was conducted in the Department of Obstetrics & Gynaecology, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka between June 2012 to July 2013. A total of 102 uncomplicated pregnant women attending at the Obstetrics & Gynaecology Department of BSMMU, Dhaka were the study population. Plasma vitamin B_{12} was measured by Abbott Axsym System using a Enzyme Immuno Assay Technique with the blood collected in a plain test tube. A serum vitamin B_{12} level of < 200 pg/mL was considered as low serum vitamin B_{12} level. The outcome variables were birth weight, small-for-date, neural tube defects and other congenital malformations.

Result: The selected pregnant women were housewife (74.5%) from low socioeconomic strata. They were generally urban resident (90.2%) with mean age being 26.4 years. In terms of education 17.6% were primary level, 30.4% SSC level, 31.4% HSC level and 20.6% graduate and higher level educated. Majority (90.2%) was urban resident with average monthly family income being Taka 29460. One-third (33.3%) was overweight and 6.9% obese. Two-thirds (66.7%) were at 37-39 weeks of gestation and nullipara. More than 60% received ANC (antenatal care) aregularly. Over one-third (35.3%) had low serum vitamin B_{12} (< 200 pg/mL). The food behaviour of the women was found to be associated with vitamin B_{12} deficiency. The pregnant women with low serum vitamin B_{12} (< 200 pg/ml) had significantly lower weekly consumption of fishes and eggs which are the rich source of vitamin B_{12}. The weekly fish, eggs and amount of milk consumed by the pregnant women with low serum vitamin B_{12} were much lower than those consumed by the pregnant women with normal serum vitamin B_{12}. The pregnant women with low serum vitamin B_{12} were more likely to carry a higher risk of adverse perinatal outcome. The incidences of low-birth weight and small-for-dates were staggeringly higher in women with low serum vitamin B_{12} (33.3% and 27.8% respectively) compared to those in women with normal serum vitamin B_{12}.

Conclusion: The study concluded that pregnant women with low serum vitamin B_{12} are accustomed to taking low fish and eggs in their daily diet compared to those who have normal serum vitamin B_{12}. The incidence of low birth weight and small-for-date babies are more prevalent in the pregnant women with subclinical deficiency of vitamin B_{12}.

Key words: Food behaviour, pregnant women, serum vitamin B_{12} and neonatal outcome etc.

Authors' information:

[1] ***Dr. Rowshan Afrooz,*** *MBBS, MS (Obstetrics & Gynaecology), Assistant Professor, Obstetrics & Gynaecology, Nightingle Medical College, Dhaka*

[2] ***Dr. Lipika Ghosh****, MBBS, MS (Obstetrics & Gynaecology), Assistant Professor, Colonel Malek Medical College, Manikganj*

[3] ***Dr. Abul Kalam Md. Faruq****, BDS, MS (Oral & Maxillofacial Surgery), Assistant Professor, Oral & Maxillofacial Surgery, Dhaka Dental College, Dhaka.*

[4] ***Dr. Mitheel-Ibna Islam****, MBBS, FCPS (Obstetrics & Gynaecology), DGO, Medical Officer, 250 Bed Shahid Shaikh Abu Naser Specialist Hospital, Khulna.*

Correspondence:* Dr. Rowshan Afrooz, Cell Phone: +88 01823883265, E-mail: dr.akmfaruq@yahoo.com*

INTRODUCTION:

Vitamin B_{12} deficiency is emerging as a public health concern in many developing countries. A progressive reduction in plasma vitamin B_{12} levels has been documented to occur during pregnancy despite maintenance of normal dietary intake of this vitamin.[1,2] A review of studies in Latin America revealed that it occurs in 40% of women and their children due to low intake of animal sources of food.[3] All the vitamin B_{12} in nature is produced by microorganisms and it is found only in foods of animal origin and vegetables contaminated with vitamin B_{12}-synthesizing bacteria.[4]

Vitamin B_{12} is an essential nutrient in the diet of humans due to its important function as a coenzyme in the remethylation of homocysteine to methionine and in the conversion of methylmalonyl-CoA to succinal-CoA. It is involved in fatty acid degradation and protein and DNA synthesis.[5] Deficiency of this nutrient is known to be associated with signs of demyelination, usually in the spinal cord. Lack of vitamin B_{12} in the maternal diet during pregnancy has been shown to cause severe retardation of myelination in the nervous system of the fetus.[6] Inadequate vitamin B_{12} status during pregnancy has consequences for both the mother and the fetus. Vitamin B_{12} deficiency is associated with increased risk for early recurrent abortion[7] and preeclampsia in pregnant women[8] and neural tube defects in infants.[9-11] A tripling in the risk for neural tube defects was reported by Ray and coworkers for infants born to women in the lowest quartile of vitamin B_{12} status compared with the highest quartile.[12]

In contrast to other water-soluble vitamins, significant amounts of vitamin B_{12} are stored in the body. As a result, it may take several years for the clinical symptoms of B_{12} deficiency to develop. Nevertheless, marginal vitamin B_{12} status in pregnancy is of concern since the vitamin B_{12} status of the mothers is associated with their fetal outcome.[13]

There are degrees of vitamin B_{12} deficiency. Overt clinical B_{12} deficiency, the more serious form, is marked by haematological or neurological abnormalities, or both, and requires prompt medical treatment. However, clinically evident deficiency accounts for less than 10% of low vitamin B_{12} concentrations observed in most surveys.[14] Subclinical vitamin B_{12} deficiency, the more common situation, is a state in which there is metabolic evidence of deficiency in a non-symptomatic patient, such as an elevated serum level of methylmelonic acid or homocysteine.[15] The main cause of subclinical vitamin B_{12} deficiency is a chronic low intake of dietary vitamin B_{12} and may not require medical intervention since it does not usually progress to clinical deficiency.[14]

As foods of animal origin, such as meat, milk and poultry are the main sources of dietary vitamin B_{12},[16] knowledge concerning the vitamin B_{12} status of pregnant women is of particular importance in parts of the world where dietary vitamin B_{12} is often low in foods from animal sources providing only marginal amounts of vitamin B_{12}.[17] As Bangladesh is one of those parts of the world, where majority of people's daily dietary intake is poor in animal sources of foods leading them to suffer from vitamin B_{12} deficiency, it is of utmost importance to study the level of vitamin B_{12} status in pregnant women and its association with their food behaviour.

METHODS:

The present cross-sectional study was carried out in Department of Obstetrics & Gynaecology, BSMMU Hospital, Dhaka, over a period of 1 year between July 2012 to June 2013. Pregnant women attending at the above mentioned Hospital were the study population. A total of 102 uncomplicated pregnant women at their third trimester who had previously booked (registered) with the antenatal clinic were included in the study. However, pregnant women with clinical diagnosis of chronic illness, such as, diabetes mellitus, hypertension, heart diseases, thyroid disorders and women with multiple pregnancies, proteinuria or any other pregnancy-related complications were excluded.

Information regarding age, gravidity and parity was obtained for each subject. Weight was measured to within 0.5 kg and height was measured to within 0.25 cm, using a portable stadiometer. Gestational age was estimated using the data of the last menstrual period, early ultrasound findings and measurement of fundal height. A single blood sample of 3 ml was collected by venepuncture in a plain test-tube tube for measurement of serum vitamin B_{12}. Neonates were examined and weighted to the nearest 10 gm on a standard beam scale balance immediately after birth. Plasma vitamin B_{12} was measured by Abbott Axsym System using a Enzyme Immuno Assay Technique with the blood collected in plain test tube. A serum vitamin B_{12} level of <200 pg/mL was considered as low serum vitamin B_{12}. Data were processed and anlysed using SPSS (Statistical Package for Social Sciences), version 17. The test statistics used to analyse the data were Chi-square (χ^2) or Fisher's Exact Test and Unpaired t-Test. The data presented on categorical scale were compared between groups using Chi-square (χ^2) or Fisher's Exact Test, while the data presented on continuous scale were compared between groups using Unpaired t-Test. Level of significance was set at 0.05 and $p < 0.05$ was considered significant.

RESULTS:

The pregnant women included in the study were generally housewife (74.5%), urban resident (90.2%) with mean age being 26.4 years. In terms of education 17.6% were primary level, 30.4% SSC level, 31.4% HSC level and 20.6% graduate and higher level educated. Average monthly family income was Taka 29460.Nearly 60% of the women were of normal BMI followed by 33.3% overweight and 6.9% obese(Table I). Two-thirds (66.7%) of the pregnant women were at 37-39 weeks of gestation, 25.5% were 40-41 weeks and 7.8% were 34-36 weeks of gestation. In terms of parity 65% were nullipara, 14.7% primipara and 19.6% multipara. More than 60% received ANC regularly and 39.2% received it irregularly (Table II).

Out of 102 pregnant women, 36(35.3%) had low serum vitamin B_{12} and the rest 66(64.7%) had normal level of vitamin B_{12}. The mean serum vitamin B_{12} was 208 pg/ml and the lowest and highest values were 60 and 294.4 pg/ml respectively (Table III). There was no significant difference between pregnant women with low and normal vitamin B_{12} status in terms of their age (p =0.252). However, mean monthly income was significantly lower in the former group than that in the latter group ($p < 0.001$). Over 70% of women with low serum vitamin B_{12} status were low level educated (primary and SSC level) as opposed to women with normal vitamin B_{12} status (44%) (p = 0.049). The parity and BMI were not found to be associated serum vitamin B_{12} status of pregnant women (p = 0.970 and p = 0.124 respectively) (Table IV).The median fish day in a week was observed to be lower in women with low vitamin B_{12} status than that in women with normal vitamin B_{12} ($p < 0.001$ and $p < 0.001$). The mean number of eggs and amount of milk consumed in a week were also less in pregnant women with low vitamin B_{12} status than that in women with normal vitamin B_{12} level ($p < 0.001$ & $p = 0.001$ respectively). However, median meat days in a week was almost similar between the groups (p = 0.301) (Table V).

The prevalence of low birth weight was significantly higher in women with low serum vitamin B_{12} than that in women with normal vitamin B_{12} level ($p < 0.001$). The proportion of small-for-date was also much higher in the former group (p = 0.002). Neural tube defects and congenital malformation were no different between the groups (p = 0.133 and p = 0.279 respectively) (Table VI).

TABLE I. Distribution of respondents by their sociodemographic features (n = 102)

Sociodemographic features	Frequency	Percentage
Age* (years)		
< 20	12	11.8
20 – 30	60	58.8
≥ 30	30	29.4
Maternal occupation		
Service	26	25.5
Housewife	76	74.5
Education		
Primary	18	17.6
SSC	31	30.4
HSC	32	31.4
Graduate plus	21	20.6
Residence		
Urban	92	90.2
Rural	10	9.8
Monthly Income (Taka)**		
< 10000	4	3.9
10000 – 20000	35	34.4
20000 – 30000	25	34.4
≥ 30000	38	37.3
BMI# (kg/m2)		
18.5 – 24.9 (Normal)	61	59.8
25 – 29.9 (Overweight)	34	33.3
≥ 30 (Obese)	7	6.9

***Mean age** = 26.4 ± 5.5 yrs; ****Mean income** = 29460 ± 2180 Taka;
#**Mean BMI** = 25.4 ± 3.4 yrs.

TABLE II. Distribution of respondents by obstetric characteristics (n=102)

Obstetric characteristics	Frequency	Percentage
Gestation age (weeks)		
34 – 36	8	7.8
37 – 39	68	66.7
40 – 41	26	25.5
Parity		
Nullipara	67	65.7
Primipara	15	14.7
Multipara	20	19.6
ANC		
Regular	62	60.8
Irregular	40	39.2

TABLE III. Distribution of respondents by serum vitamin B12 status (n = 102)

Serum vitamin B12	Frequency	Percentage
< 200	36	35.3
≥ 200	66	64.7

***Mean serum vitamin** = 208.0 ± 50.7 pg/ml; **range** = 60 – 294.4 pg/ml

TABLE IV. Comparison of demographic and obstetric characteristics between groups

Demographic features	Serum vitamin B12 (pg/ml)		p-value
	< 200 (n = 36)	≥ 200 (n = 66)	
Age* (yrs)	27.2 ± 4.3	25.9 ± 6.1	0.252
Monthly family income* (Taka)	19375 ± 1703	35967 ± 3162	< 0.001
Education#			
Primary	14(38.9)	17(25.8)	0.049
SSC	12(33.4)	12(18.2)	
HSC	6(16.7)	22(33.3)	
Graduate plus	4(11.0)	15(22.7)	
Parity#			
Nullipara	23(63.9)	44(66.7)	0.970
Primipara	6(16.7)	9(13.6)	
Multipara	7(19.5)	13(19.7)	
BMI* (kg)	24.8 ± 3.4	25.9 ± 3.4	0.124

Figures in the parentheses denote corresponding percentage;
Data were analysed using χ^2 **Test.**
*Data were analysed using **Unpaired t-Test** and were presented as **mean ± SD.**

TABLE V. Association between food behaviour and serum vitamin B12

Food behaviour-related variables	Serum vitamin B12		p-value
	< 200 (n = 36)	≥ 200 (n = 66)	
Median fish days in a week*	2.1 ± 0.9	3.3 ± 1.7	< 0.001
Median meat days in a week*	1.9 ± 0.3	2.2 ± 0.2	0.301
Mean number of eggs taken in a week*	1 ± 1	3 ± 1	< 0.001
Mean amount of milk in a week* (liter)	0.33 ± 0.13	0.88 ± 0.11	0.001

Figures in the parentheses denote corresponding percentage;
* Data were analysed using **Unpaired t-Test** and were presented as **mean ± SD.**

TABLE VI. Association between maternal serum V-B12 and perinatal outcome

Outcome variables	Serum Vitamin B12 (p g/ml)		p-value
	< 200 (n = 36)	≥ 200 (n = 66)	
Birth weight (kg)			
< 2.5	12(33.3)	4(6.1)	< 0.001
≥ 2.5	24(66.7)	62(93.3)	
Mean ± SD*	2.6 ± 0.4	3.1 ± 0.7	
Small-for-date#	10(27.8)	4(6.1)	0.002
Neural tube defect**	2(5.6)	0(0.0)	0.133
Other congenital malformation**	2(5.6)	1(1.5)	0.279

Figures in the parentheses denote corresponding percentage;
*Data were analysed using **Unpaired t-Test** and were presented as **mean ± SD.**
#Data were analysed using χ^2 **Test;**
Data were analysed using **Fisher's Exact Test

ORIGINAL ARTICLE

DISCUSSION:

The present study demonstrated that subclinical vitamin B_{12} deficiency of pregnant women is associated with their food behaviour and carries a higher risk of adverse perinatal outcome. The pregnant women with low serum vitamin B_{12} (< 200 pg/ml) had significantly lower weekly consumption of fishes and eggs which are the rich source of vitamin B_{12}. The weekly fish consumption of pregnant women with low serum vitamin B_{12} was on an average 1 day less than the pregnant women with normal serum vitamin B_{12}. The average number of eggs taken weekly by the former group was less than that taken by the latter group. The amount of milk taken by the pregnant women with low serum vitamin B_{12} was drastically lower than their counterparts with normal serum vitamin B_{12} level. Of 102 pregnant women, 36 (35.3%) had serum vitamin B_{12}< 200 pg/mL, which is higher than a recent study conducted on non-pregnant Bangladeshi women (22%) (Survey on Micronutrients, 2013)[18] but much lower than that found in a study conducted on non-pregnant Jordanian women (67%). Guerra-shinohara and colleagues (2004) have described a 37.3% decrease in vitamin B_{12} levels from the beginning to the end of pregnancy in Brazilian women. This fall is thought to be physiological, owing to an increase in plasma volume and a change in hormonal status[19] and an increase in the requirements of the vitamin, secondary to pregnancy.[20] In a study of nutritional factors related to anaemia in pregnant women attending an antenatal clinic in Nigeria,[21] 9% of pregnant women had serum vitamin B_{12} levels below the lower end of the normal range. Chéry and colleagues[22] found that low vitamin B_{12} levels in the third trimester of pregnancy correlated with a rise in plasma total homocysteine during this time and suggested that subclinical deficiency may start to appear at this stage, particularly in mothers on diets deficient in animal foods. As inadequate vitamin B_{12} increases the risk of adverse maternal and foetal outcome,[8,23] over one-third of the pregnant women in our study were at increased risk of adverse perinatal outcome.

The present study also observed a significantly inverse association of income and education with vitamin B_{12} level with higher the income and level of education lower is the probability of having low serum vitamin B_{12}. Muthayya and associates[24] suggest that a low educational level, poor anthropometric status in early pregnancy, low weight gain in the second trimester of pregnancy & a low serum vitamin B_{12} concentration throughout pregnancy are significant determinants of IUGR (Intrauterine Growth Restriction) in urban Indian women. The inverse relationship between maternal educational level and risk of IUGR persists even after adjusting for maternal age, parity and maternal weight in early pregnancy. Several other studies also reported maternal illiteracy and low socioeconomic status to be the major risk factors for IUGR.[25,26]

In the present study, the perinatal outcome of pregnant women with low serum vitamin B_{12} was unfavourable in terms of birth weight and small-for-date babies. Low serum vitamin B_{12} was found in 15.6% of the cases, which is much lower than the published national figure (36%).[27] This might be due to tertiary hospital-based study, which may not be the true representative of the community population. However, analyses of association between maternal serum vitamin B_{12} and perinatal outcome revealed a significantly lower mean birth weight (on an average 0.5 kg less) and a higher incidence of small-for-date neonates (on an average 22% higher) in pregnant women with low serum vitamin B_{12} compared to those in normal serum vitamin B_{12} level.

Muthayya and colleagues[24] demonstrated that women in the lowest tertile for vitamin B_{12} concentration during each of the three trimesters of pregnancy had significantly higher risk of IUGR (adjusted OR=5.98). They also reported that better socioeconomic conditions and improved nutritional status well-correlates with vitamin B_{12} status during pregnancy, which in turn, reduces the incidence of IUGR. An earlier study reported negative correlations between birth weight and maternal vitamin B_{12} levels at delivery in smokers in Western women,[28] although present study did not investigate smoking habit of the selected women, which might act as a confounder. Thus, the findings suggest that early detection of vitamin B_{12} deficiency in pregnancy followed by appropriate intervention are likely to play an important role in reducing IUGR which contributes substantially to low birth weight & small-for-dates. Molloy & associates[29] demonstrated that starting pregnancy with an inadequate vitamin B_{12} status may increase the risk of birth defects such as NTD, and may contribute to preterm delivery, although NTD in the present study was not significantly associated with women of low serum vitamin B_{12} level.

Nutritional status during pregnancy has a direct influence upon birth weight. Adequate supply of micronutrients is known to be very important in pregnancy and there is much evidence to support a role of folate in early embryonic development.[30] A clear causal association has been shown between reduced maternal folate and neural tube defects[31,32] and other congenital malformations[33] as well as early pregnancy loss.[34] More recently Rao et al[35] observed a positive association between infant birth weight and dietary intake of folate-rich foods in rural Indian women. So maternal diet during pregnancy is highly important for the health of the fetus.[35]

Vitamin B_{12} plays an integral role in folate-dependent homocysteine metabolism[36] as a rate-limiting co-factor in the conversion of homocysteine to methionine and a limited supply of vitamin B_{12} during pregnancy may have adverse consequences on fetal growth. Furthermore, low serum levels of maternal vitamin B_{12} have been associated with risk of neural tube defect-affected pregnancy.[37,38] Vitamin B_{12} is known to be a significant predictor of tHcy levels in neonates, with relatively greater importance in the first few months of life, before folate assumes primacy as the predictor of tHcy (total serum homocysteine). A deficiency of either vitamin B_{12} and/or folic acid is likely to affect homocystine metabolism, resulting in an elevation of plasma homocystine with relatively low methionine level. During pregnancy an increase Hcy (Homocysteine) level has been shown to be a risk factor for IUGR, intrauterine death, neural tube defects and low birth weight. Murphy and associates[39] have reported that mothers in the highest total Hcy tertile at 8 weeks of pregnancy had three times and at labour nearly four times the odds of giving birth to a neonate in the lowest birth weight tertile. The role of vitamin B_{12} in pregnancy outcome has been explored by Molloy et al.[29] who suggested that vitamin B_{12} status may be an important factor in maintaining optimal tHcy during pregnancy, although the potential role of vitamin B_{12} in the determination of birth weight has yet to be explored. Therefore, additional studies are needed to elucidate the role of vitamin B_{12} together with Hcy in the prevention of adverse outcomes.

The present study had several limitations which deserve mention so that future studies on the same issue could overcome the limitations faced in the present one.

LIMITATIONS:

1. The food behaviour of the women was assessed by 7-days dietary recall, which might be inherently associated with recall bias.

2. As studies by other researchers have shown that folate & homocystine are also associated with maternal and perinatal outcome, it cannot be over-emphasized that the adverse perinatal outcome attained in the pregnant women have had occurred solely due to low serum vitamin B_{12}.

CONCLUSION:

From the findings of the study it can be concluded that over one-third of the Bangladeshi pregnant women possess low vitamin B_{12}. The pregnant women with low serum vitamin B_{12} are used to taking low fish and eggs in their daily diet

ORIGINAL ARTICLE

compared to those who have normal serum vitamin B_{12}. The milk consumption of the pregnant women are also much lower than their counterparts with normal serum vitamin B_{12} level. The incidence of low birth weight and small-for-date babies are more prevalent in the pregnant women with subclinical deficiency of vitamin B_{12}. So a well-correlation is observed between pregnant women of low-socioeconomic group and their food-behaviour, between food-behaviour and their serum vitamin B_{12} status and vitamin B_{12} status during pregnancy and perinatal outcome.

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