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**Original Research Article**

# Screening and prevalence of vitamin B12 deficiency among pregnant women

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## ABSTRACT

**Background:** Vitamin B12 deficiency (serum vitamin B12 <148pmol/L) is considered to be an important cause of anaemia in pregnancy. Pregnant women with this deficiency have an increased risk of developing preeclampsia, intra-uterine growth retardation and preterm labour. The current study was undertaken to determine the prevalence of vitamin B12 deficiency in pregnant women and its association with their dietary habits and socioeconomic status.
**Methods:** This was a cross sectional study conducted in a tertiary care institute over a period of 2 months. Pregnant women visiting antenatal OPD were included in the study and those receiving vitamin B12 supplements, antacids, antiepileptic medication or methotrexate were excluded. Each participant was subjected to a predesigned structured questionnaire and their serum vitamin B12 concentration was done using quantitative determination by microplate enzyme immunoassay.
**Results:** Of the 97 women included, majority were in the age group 20-25years (57.73%). 87.62% (85 women) were found to have anaemia and a total of 44women (45.36%) had vitamin B12 deficiency.
**Conclusions:** The prevalence of vitamin B12 deficiency in north India is considerably high, which may be the cause of neural tube defects and poor birth outcomes and neurological deficit in children born to these mothers.

**Keywords:** Pregnancy, Vitamin B12 deficiency, Pregnant women

## INTRODUCTION

WHO defines anaemia in pregnancy as haemoglobin concentration of <11 gm/dl. It is a public health problem especially in developing countries and is associated with adverse perinatal outcomes. It is considered to be of public health significance if its prevalence is 5% or higher in population studies and severe public health problem if >40%. In India, 52% pregnant women and 58% lactating women have anaemia as per National Family Health Survey 5 (2019-21).

Vitamin B12 deficiency (serum vitamin B12 <148pmol/L) is considered to be an important cause of anaemia in pregnancy.[1] Vitamin B12, also known as "cobalamin" is a water-soluble vitamin is present in food products of animal origin such as fish, meat, eggs and dairy products. In fortified food and supplements, the synthetic form of vitamin B12, known as cyanocobalamin is used. It plays an important role in DNA synthesis, regulation and also in fatty acid synthesis and energy production. Vitamin B12 also acts as a coenzyme in folate synthesis, therefore it is crucial for erythropoiesis and normal neurodevelopment.

Vegetarians and vegans are considered to be at high risk for developing a vitamin B12 deficiency. Numerous studies on smaller population groups confirmed that both vitamin B12 intake and serum vitamin B12 concentrations increase progressively from vegans to lacto-ovo-vegetarians, to those who consume fish or some meat, to omnivores. Deficiency of vitamin B12 is not confined to these groups and can occur in any population.[2] Vitamin B12 deficiency poses an increased risk of developing hypertensive pregnancy disorders, growth restriction and

preterm labour.[3,4] Recently, association between neural tube defects and low vitamin B12 status in mothers have also been found. Hence, the risk for birth defects increases when pregnancy starts with a deficient or inadequate vitamin B12 status, although this association clearly does not prove any causal relationship.[3] Studies are also suggestive of an association of B12 deficiency in pregnancy with increased adiposity which might lead to insulin resistance and gestational diabetes mellitus.[5] Children of mothers with vitamin B12 deficiency are also at risk for low birth weight.[6,7]

Studies have not yet established the best cut off for maternal serum vitamin B12 levels before conception, during pregnancy and in the lactational period. Also, the racial/ethnic differences in the serum levels have not been studied. The prevalence of vitamin B12 deficiency among north Indian pregnant women is not known and its association with modifiable factors is not clearly defined. The current study was undertaken to determine the prevalence of vitamin B12 deficiency in pregnant women and its association with their dietary habits and socioeconomic status through a pre designed structured questionnaire.

## METHODS

The current study was a cross sectional study conducted in a tertiary care institute over a period of 2 months, April 2019 to May 2019 after approval by institutional ethics committee.

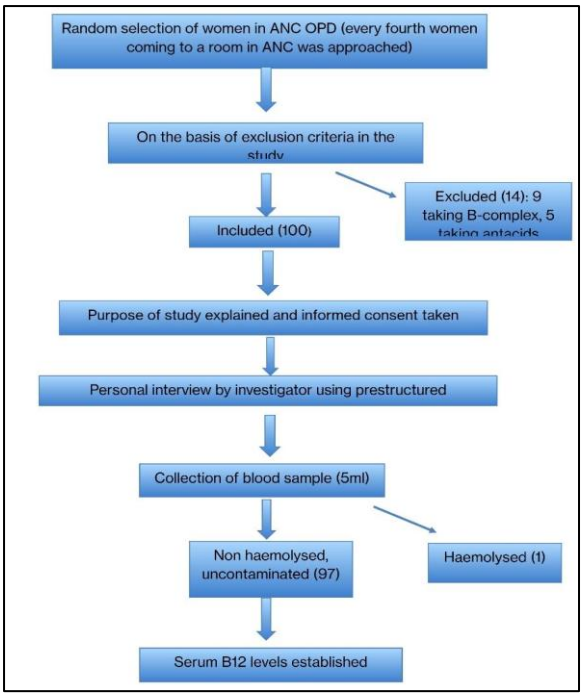


**Figure 1: Methodology.**

Pregnant women visiting antenatal OPD were included in the study after written informed consent. Those receiving vitamin B12 supplements, antacids, antiepileptic medication or methotrexate were excluded. Each participant was subjected to a predesigned structured questionnaire and their serum vitamin B12 concentration was done using quantitative determination by microplate enzyme immunoassay. Blood sample (5ml) was collected under stringent aseptic conditions in a plain vacutainer. The sample was centrifuged and the serum was separated. This was stored at -20 degree Celsius till the tests were applied. Quantitative estimation of serum B12 was done using a microplate enzyme immunoassay in Department of Biochemistry, VMMC & SJH (Figure 1). The questionnaire was analysed using IBM-SPSS software. Associations were tested using chi square test. p value of <0.05 was considered significant.

## RESULTS

Of the 97 women included in the study, majority were in the age group 20-25years (57.73%). 87.62% (85 women) were found to have anaemia and a total of 44women (45.36%) had vitamin B12 deficiency (Table 1).

**Table 1: Sociodemographic characteristics.**

| Variables | Frequency (n) | B12 deficiency (number) |
|---|---|---|
| **Total** | 97 | 44 (45.36%) |
| **Age of patients** | | |
| <20 | 5 (5.15%) | 2 (40%) |
| 20-25 | 56 (57.73%) | 30 (53.57%) |
| 26-30 | 31 (31.95%) | 9 (29.03%) |
| 31-35 | 5 (5.15%) | (60%) |
| **Trimester** | | |
| 1st | 8 (8.28%) | 3 (37.5%) |
| 2nd | 48 (47.42%) | 22 (47.82%) |
| 3rd | 43 (44.32%) | 19 (44.18%) |
| **Socio economic status** | | |
| Upper (26-29) | 12 (12.37%) | 4 (33.33%) |
| Upper middle (16-25) | 20 (20.16%) | 11 (55%) |
| Lower middle (11-15) | 42 (43.29%) | 19 (45.23%) |
| Upper lower (5-10) | 22 (22.68%) | 9 (40.9%) |
| Lower (<5) | 1 (1.03%) | 1 (100%) |
| **Working status** | | |
| Housewife | 85 (87.62%) | 39 (45.88%) |
| Outside home | 12 (12.37%) | 5 (41.66%) |
| **Educational status** | | |
| Illiterate | 14 (14.33%) | 7 (50%) |
| Senior secondary | 62 (63.91%) | 31 (50%) |
| Graduate and above | 21 (21.64%) | 6 (28.57%) |
| **Anaemia state** | | |
| None | 12 (12.37%) | 5 (41.66%) |
| Mild | 20 (20.17%) | 10 (50%) |
| Moderate | 62 (63.91%) | 29 (46.77%) |
| Severe | (33.09%) | 0 |
| Total anaemia | 85 (87.62%) | 39 (88.63%) |

**Table 2: Serum vitamin B12 levels.**

| Serum vitamin B12 levels (pg/ml) | Number of patients (n=97) | Percentage |
|---|---|---|
| **≤ 100** | 2 | 2.1 |
| **101 - 150** | 9 | 9.3 |
| **151-200** | 33 | 34 |
| **≥200** | 53 | 54.8 |

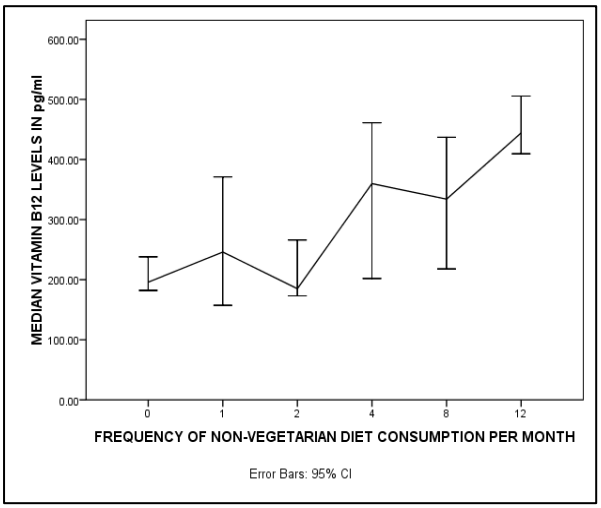


**Figure 2: Median vitamin B12 levels of pregnant women with respect to their frequency of meat consumption.**

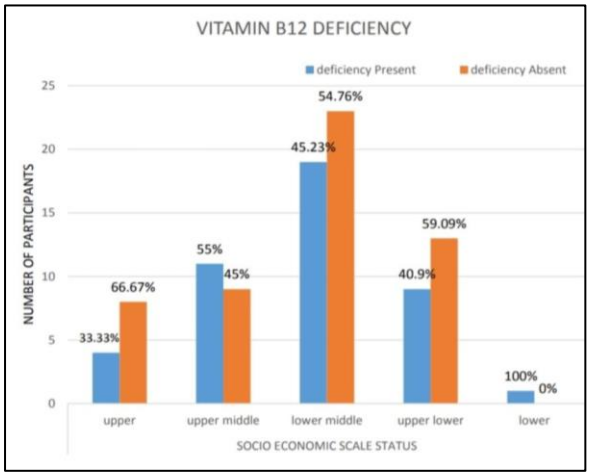


**Figure 3: Prevalence of vitamin B12 deficiency with respect to the socioeconomic status.**

Anaemia was classified into mild, moderate and severe according to ICMR guidelines. Socioeconomic status was calculated using modified Kuppuswamay socioeconomic scale.

Normal range for serum vitamin B12 is 200 pg/ml - 835 pg/ml. Vitamin B12 deficiency (<200 pg/ml) was found in 45.36% of participants, of which mostly (34%) had serum B12 ranging from 151 pg/ml - 200pg/ml and 9.3% from 101 pg/ml-150 pg/ml, whereas only 2.1% women had levels below 100 pg/ml. 54.6% women had serum vitamin B12 levels more than 200 pg/ml (Table 2).

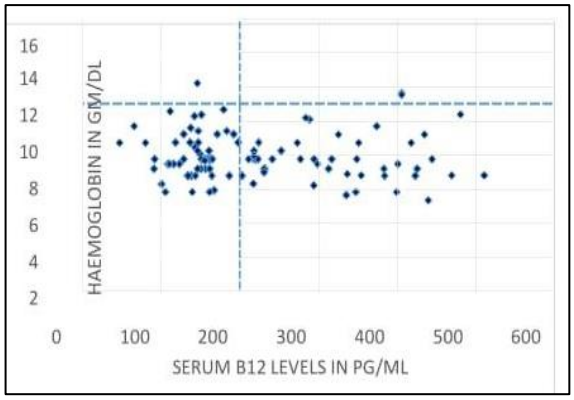


**Figure 4: Correlation of Serum Vit B12 levels with haemoglobin in pregnant women.**

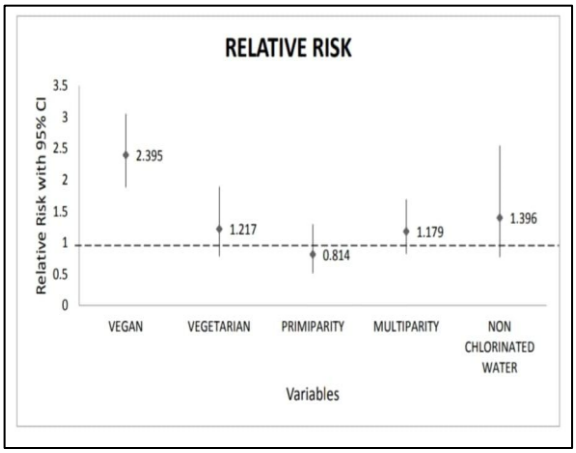


**Figure 5: Relative risk of various factors affecting vitamin B12 levels in pregnancy.**

Serum vitamin B12 levels were found to be gradually rising with increase in frequency of meat consumption. Levels were highest among the women who were consuming meat 12 times a month. Hence meat consumption was found to be one of the chief factors affecting vitamin B12 level (Figure 2).

Vitamin B12 deficiency was present in all strata of society on the basis of socioeconomic classification, but the graph clearly indicates a higher prevalence of vitamin B12 deficiency in patients belonging to lower and upper middle class (Figure 3).

There was clustering of cases in the region with serum B12 less than 200 pg/ml and haemoglobin levels less than 11 g/dl indicating the high occurrence of vitamin B12 deficiency in anaemic women. It also shows high prevalence of anaemia during pregnancy (Figure 4).

Pregnant women who were having a vegan diet have a relative risk of 2.395 as compared to non-vegan participants and there is a significant association between these two with a p value of 0.007 which is ≤ 0.05. Primiparity was not found to be associated with vitamin B12 deficiency whereas vegetarian diet, multiparity and intake of non-chlorinated water are associated with vitamin B12 deficiency (Figure 5).

It was found that 10.6% (n= 7) of the non-vegetarian women sincerely believed that diet consisting of meat during pregnancy is harmful to the foetus and may induce abortion whereas 89.4% (59) women had no such beliefs. 4.54% (3) said they avoided chicken because it was not palatable. 12.9% (4) of the vegetarian women said they avoided milk and milk products as they were unable to tolerate them because caused nausea and vomiting. 29.03% vegetarian women preferred milk and milk products. 6.55% non-vegetarian pregnant females preferred meat over other food items and had a high frequency of meat consumption.

## DISCUSSION

Vitamin B12 (a water-soluble vitamin) helps in red cell synthesis and normal functioning of brain and nervous system.[8] This along with folic acid is needed for methionine formation from homocysteine, which in turn is important for DNA synthesis. Main source of vitamin B12 is of animal origin.[10] It is also present in fermented foods and uncooked plant-based food contaminated with vitamin b12 producing bacteria or algae.[11] Gastrointestinal disorders, malabsorption, drugs like metformin, low levels of vitamin B12 binding proteins and proton pump inhibitors also lead to its deficient state. Vegetarians, women from low socio-economic strata and those refusing to consume meat due to social or religious reasons are at increased risk.[11,13-15]

Pregnancy is a critical time period associated with increased physiological demand for various micronutrients and macronutrients. A dietary intake of 1 mcg/day for adults and 1.5 and 1.2 mcg/day for pregnancy and lactation respectively is recommended by the Indian Council of Medical Research. National institute of heath recommends that pregnant and lactating women should consume 2.8 mcg of vitamin B12 per day. In the current study, we found that, among the 114 women, only 9 (7.89%) were consuming B12 supplements.

Among the 97 participants, 6 were consuming exclusively vegan diet and all of them had B12 levels below 200 pg/ml. There was significant association between vegan diet and Vitamin B12 deficiency with a p value of 0.007 which is ≤0.05.

Prevalence of anaemia in pregnant women worldwide is 40% as per WHO and it is 50% in India as per NFHS 4. In the current study, the prevalence of anaemia was found to be 87.62% and 67% for moderate to severe anaemia which is higher than the national data. This is because, ours was a hospital-based study wherein most of the women (91.74%) were in late pregnancy (second or third trimester) which is associated with haemodilution and thus decreased haemoglobin levels. Moreover, as the study was conducted in a tertiary care hospital most where most of the patients are referred from the peripheral heath care centres.

In addition to high prevalence of anaemia, there was high prevalence of Vitamin B12 deficiency (<200pg/ml) as well (45.36%). The results were similar to study from a semiurban area in Bangalore, with a prevalence of 51% and in Pune with a prevalence of 80% and 65% among rural and urban pregnant women respectively.[16,17]

In this study 68% of the women were non-vegetarians, there was a high prevalence (42.4% with vitamin B12 levels <200mg/dl) of vitamin B12 deficiency among them and 32% were vegetarian of which 51.6% had vitamin B12 deficiency, which is in accordance to the study done by Rashid et al where they found that the pregnant and lactating women on vegetarian and vegan diet were at high risk of deficiency due to increased metabolic demand of vitamin b12.[18] The higher prevalence among the nonvegetarian women can be explained as most of them belong to the low socio-economic strata (67% belonging to lower middle class and below) having low meat consumption.

Vitamin B12 deficiency was associated with low socio-economic status in few previous studies, whereas in the current study no significant association was found between socioeconomic status and serum vitamin B12 levels as it was prevalent in all socioeconomic classes with slightly more prevalence in low and upper middle class. The probable reason for a higher prevalence in low socioeconomic class is due to their inability to afford non-vegetarian food, whereas in upper middle class most of the study population belonged to Hindu religion (95%) who prefer vegetarian diet due to religious beliefs and there is ignorance regarding vitamin B12 and balanced diet.

The present study is representative of the women residing in north India as very few studies have been conducted in this part of the country, but the limitation is its small sample size. Further research should be conducted on a large scale. These findings can have far reaching implications as the high-risk groups can be advised high dietary intake of food products of animal origin or vitamin B12 supplementation, thus improving the quality of antenatal care and foetal outcome.

## CONCLUSION

Vitamin B12 deficiency in north India is considerably high, which may be the cause of neural tube defects and poor birth outcomes and neurological deficit in children born to these mothers. There is a very little awareness about vitamin B12 and its importance among the pregnant

women. The common factors affecting vitamin B12 status are mostly modifiable, such as change in dietary habits, preventing contamination of food and drinking water. Knowledge in this regard should be provided through community-based awareness programmes and group discussions to all pregnant women. There is a need to educate and motivate women to translate knowledge into practice, involve the elders in the family or the community who are often the decision makers and also address social and cultural barriers and misconceptions prevailing among them.

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