



# Study of Serum Vitamin B12 Level in Population Consuming Tobacco in Various Forms as Compared to Non-Tobacco Users

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## ABSTRACT

**Introduction:** Tobacco consumption is risk factor for so many diseases. Experimental and clinical evidence both suggest that there is some connection between metabolism of vitamin B12 and that of cyanide. Smoking releases cyanide and other toxins.

**Methods:** In this study out of 120 patients 40 were smokers, 40 were tobacco chewers and 40 were nontobacco users in any forms (controls). Prospective case control study was done. Measurement of serum Vitamin B12 level was done. The patients having Vitamin B12 level <210pg/ml were considered deficient and those having >210pg/ml were considered normal. Serum Vitamin B12 level was done using radio immune-assay by Immulite analyzer.

**Results:** It was seen that in tobacco consuming patients vitamin B12 levels in serum were low. The 69% patients with vegetarian diet and 37% patients with mixed diet were having Vitamin B12 deficiency. Odds ratio of 3.87 is suggesting that Vitamin B12 deficiency was quite common in Vegetarian diet.

**Conclusions:** The association between tobacco use and vitamin B12 deficiency was found statistically significant. So correlation can be there. However large study required.

**Key words:** Vitamin B12, Smoking, Tobacco Chewers

## INTRODUCTION

Tobacco use is highly prevalent across the world. It kills millions of people worldwide causing immense suffering and also has enormous economic and social cost.<sup>1</sup> Worldwide, tobacco use causes nearly 6 million annual deaths, and current trends show that it will cause more than 8 million deaths annually by 2030.<sup>2</sup> Although there is wide documentation of the adverse effects of tobacco smoking on variety of diseases and disturbances, but effects of tobacco use on Vitamin B12 is studied less.<sup>3</sup> Tobacco use is highly prevalent in India with smoking and chewing being two most common forms of use.<sup>4</sup> The estimates of the Global Adult Tobacco Survey (GATS) conducted among persons 15 years of age or older during 2009-10 indicate that 34.6% of the adults (47.9% males and 20.3% females) are current tobacco users.<sup>4</sup> Smoking affects both vitamin b12 and folic acid levels in body.<sup>5</sup>

Smokeless tobacco has also been shown to affect the immune response both in vitro and in vivo.<sup>6</sup>

Vit B12 contributes to detoxification of exogenous cyanide, which is derived from tobacco smoke.<sup>7</sup>

Optic neuritis in smokers is related to deficiency of hydroxocobalamin form of vitamin b12. In pipe smokers, due to high amount of cyanide, a good amount of hydroxocobalamin gets converted into cyanocobalamin.<sup>7</sup>

Components of tobacco smoke contain high amount of cyanide which adversely affects b12 metabolism. It is found that smokers have low levels of vitamin B12 in serum with increased excretion of B12 in urine.<sup>8</sup> In addition to b12, chronic smoking also affects folate levels in human body.<sup>9</sup> Serum folic acid concentrations are found to be lower in smokers as compared to non-smokers.<sup>9</sup>

The UK Vegan Society, the Vegetarian Resource Group, and the Physicians Committee for Responsible Medicine, among others, deny that non-animal food sources of vitamin B<sub>12</sub> are reliable and recommend that every vegan should consume food that is fortified with B<sub>12</sub>.<sup>10,11,12</sup>

The prevalence of vitamin B12 deficiency is higher among vegetarians due to suboptimal intake. Therefore they should take regular supplements.<sup>13</sup>

Here, in this study, our objective is to find out if there is any significant association between tobacco consumption and vitamin b12 levels in serum. We will also try to look for association between dietary pattern and serum b12 levels.

### MATERIALS AND METHODS

The present study was carried out in OPD and indoor patients of our institute from July 2014 to August 2016. The study group included 120 patients. Among them 40 were smokers (GROUP A), 40 were tobacco chewers (GROUP B) and 40 were nontobacco users (control/GROUP C). Informed consent was obtained before enrolment of patients. In Group A, the cases who smoked daily for >/+ 6 months were considered smokers; in Group B, the cases who chewed tobacco or consumed gutkha daily for >/= 6 months were considered as tobacco chewers; in Group C, those who have never smoked or consumed tobacco in their life time were considered as nonsmokers or non-tobacco users respectively, also they were free of overt disease. In all groups age was 18 to 60 years of patients. In all the groups subjects who were on B complex multivitamins, anti-S-adenosyl methionine, 6-azarudine, antropterin, tamoxifen, anti-epileptics, methotrexate, metformin were excluded, also those with diabetes mellitus, hypertension, cancer, systemic illnesses, thyroid dysfunction, renal failure rheumatoid arthritis, alcoholics were excluded. Any other causes for Vitamin B12 deficiency were excluded. The serum Vitamin B12 level was done using radio-immune assay by Immulite analyzer. The subjects were asked to fast for 10-12 hours and 10 ml of blood was collected from the anterior cubital vein after aseptic measures in sitting up position. Vitamin B 12 level <210 pg/ml was considered deficient and above that considered as normal. All the patients were examined in details for symptoms and signs of Vitamin B12 deficiency, detailed history and physical examinations were done. Detailed smoking habit and tobacco consumption history were taken.

Analysis was done using student's t test and 95% confidence interval was taken. Mean serum vitamin b12 level was compared in tobacco users and non-tobacco users; also comparison of b12 levels

was made between persons using vegetarian diet and mixed diet. P value <0.05 was to be taken as significant association. (if p value found <0.05, null hypothesis suggesting that there is no association between b12 levels and tobacco would be rejected.)

### RESULTS

It was found that in tobacco consumption groups (either in form of smoking or tobacco chewers) the Vitamin B12 level was low. The table described below indicates that in tobacco consuming groups, mean vitamin b12 levels in serum was low and absolute number of subjects having vitamin b12 deficiency was higher. Statistical analysis was made using student's t-test and the p value obtained was <0.05(95% confidence interval).

Table 1 indicated that there was significant association between tobacco consumption and vitamin b12 deficiency. P value was <0.05 for tobacco consumers v/s non tobacco users with serum b12 levels. 72.5% patients from smokers and 60% of tobacco chewers had serum b12 levels <210 pg/ml. whereas only 20% of non tobacco consumers were found to have b12 levels <210 pg/ml.

Table 2 indicated that there was significant association between vegetarian diet and vitamin b12 deficiency. As mentioned in the above table 2, 59.01% with vegetarian diet had low b12 levels compared to mixed diet in which the low levels of serum b12 were found in 40.98% only. P value obtained using chi square test was <0.05, thus proving significant association between vegetarian diet and b12 deficiency.

**Table1: Case controls and Vitamin B12 level**

| Groups          | n  | Serum Vit B12 Level (Pg/MI) (mean ± SD) | Vit B12 Deficiency* |           |
|-----------------|----|---|---------------------|-----------|
|                 |    |   | Present             | Absent    |
| Smokers         | 40 | 182.32 ± 48.56                          | 29(72.5%)           | 11(27.5%) |
| Tobacco chewers | 40 | 205.05 ± 56.05                          | 24(60%)             | 16(40%)   |
| Controls        | 40 | 244.57 ± 29.41                          | 08 (20%)            | 32(80%)   |

\*(Cut off value for vit.b12 deficiency-<210 pg/ml)  
 Student's t test applied for statistical significance  
 P value for smoker v/s control- <0.001 (<0.05)  
 P value for tobacco chewers v/s control- <0.001 (<0.05)

**Table 2: Vitamin B12 deficiency in patients with different diet pattern**

| Dietary Pattern | Vit B12 level |             | Total |
|-----------------|---------------|-------------|-------|
|                 | Normal (n=59) | Low(n=61)   |       |
| Vegetarian      | 16 (27.11%)   | 36 (59.01%) | 52    |
| Mixed           | 43 (72.89%)   | 25 (40.98%) | 68    |

Odds ratio (95% Confidence interval) 3.87 (1.79-8.34);  
 P value <0.001

## DISCUSSION

The results and observations obtained above are suggestive of significant association between tobacco consumption and vitamin b12 deficiency. It also concluded that vitamin b12 deficiency is quite common with vegetarian diet.

There was no significant difference in age of the subjects in all three groups. All other common established causes of b12 deficiency were excluded.

Our study findings correlate with a previous study of Salahuddin et al.<sup>14</sup> There were 75 smokers and 75 controls in that study. Mean vitamin b12 levels in smokers was 248.69 (+/-98.19), while mean b12 levels in controls was 427.30 (+/-139.89). In present study out of 40 smokers, mean b12 level was 182.32 and among 40 controls, mean b12 level was 244.57. The above mentioned comparison between present study and previous Salahuddin et al study points towards significant association between tobacco and vitamin b12 deficiency.

In this study we found that among 52 subjects with vegetarian diet, 36 subjects (69%) had b12 deficiency, whereas among 68 subjects with mixed diet, only 25 had vit b12 deficiency (37%). There was a study in 2012 of Andrews Liggy et al in which 225 persons out of 260 who were vegetarians had b12 deficiency (86%). In contrast only 35 out of 136 subjects with mixed diet had b12 deficiency (25%).<sup>15</sup> This observation supports our finding that vegetarians are more likely to have b12 deficiency.

Although we found significant association between tobacco use and vitamin b12 deficiency, further investigation is required to establish causal relationship between them. Power of study was also low. Because of small sample size, to extrapolate the findings to general population, large scale multi-centric studies are required. Further research is needed to find out the exact mechanism for tobacco induced vitamin b12 deficiency. A proposed mechanism that cyanide in tobacco interferes with b12 metabolism and increases its excretion<sup>8</sup>. This may be due to mechanism related to equilibrium of b12 levels between blood and tissue. In our study, we did not include urinary levels of vitamin b12. Possibility of unknown confounding factors cannot be ruled out. As tobacco users in general have poorer eating habits, nutritional factors may be contributing towards vitamin b12 deficiency. Data from the Second National Health and Nutrition Examination Survey indicates that smokers are less likely to consume fruits and vegetables, particularly those high in vitamin C and carotenes.<sup>16</sup> However, larger studies in future will help determine causal association as well as dose response relationship between tobacco and b12 levels.

## CONCLUSION

We conclude by this study that vitamin b12 deficiency was found more in tobacco consumers. We also found that vegetarian patients were at more risk of developing Vitamin B 12 deficiency as compared to patients with mixed diet. Large scale multi-centric studies are required to build a population based preventive strategy for prevention of Vitamin B12 deficiency in tobacco users.

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