Original Research Article

Evaluation of serum vitamin B12 level in patients of metabolic syndrome

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ABSTRACT

Introduction: The metabolic syndrome (MetS) is a rising public-health problem and clinical challenge worldwide. Vitamin B12 deficiency is associated with MetS. But its role as independent risk factor for MetS is not clear. Objective of our study was to evaluate serum vitamin B12 levels in MetS cases and its association with parameters of MetS

Material and Methods: Total 50 MetS cases and 50 age and sex matched healthy controls were included in the study. Cases were selected according to new international diabetes federation (IDF) criteria of MetS. Estimation of serum vitamin B12 were done on electrochemiluminescent machine immulite-1000 by electrochemiluminesce principle. Estimation of fasting blood sugar, triglyceride and HDL were done on advia 1800 chemistry autoanalyzer. Waist circumference was measured by measuring tape. Blood pressure was measured by sphygmomanometer.

Result: The median values of vitamin B12 in cases were 210 pg/ml with the interquartile difference of 293.75 and the median value of vitamin B12 in controls were 178 pg/ml with the interquartile difference of 137.5. The differences between mean ranks of two groups were not significant (P=0.25). Vitamin B12 were found to be positively correlated with serum triglyceride (correlation coefficient 0.337; P=0.001) and serum HDL level (correlation coefficient 0.207; P= 0.039).

Conclusion: There was no significant difference of serum vitamin B12 levels in metabolic syndrome patients. Serum vitamin B12 was positively correlated with serum triglyceride and HDL level.

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1. Introduction

The metabolic syndrome (MetS) is a rising public-health problem and clinical challenge worldwide. It prevalence increased with age. Main component of the MetS are central obesity, hyperglycemia, hypertension hypertriglyceridemia, and low high-density lipoprotein (HDL) cholesterol. Overweight, sedentary life, aging, lipodystrophy are risk factor of MetS. It is a common cause for development of type 2 diabetes and cardiovascular disease. It also increase susceptibility of polycystic ovary syndrome, asthma, sleep disturbances, fatty liver, cholesterol gallstones and fatty liver. MetS is more prevalent in developed countries, but its prevalence is rising day by day in developing country like India.

Vitamin B12 or cynocobalamin is a water soluble hematopoietic vitamin that helps in maturation of RBC. It act as a coenzyme for Methionine synthase and help in remethylation of homocysteine. It also play important role in cardiovascular functions. Biological reference range of serum vitamin B12 is 206-678 pg/ml. According to WHO, value less than 150 pg/ml considered as a deficiency and value more than 201 pg/ml considered as acceptable.

Vitamin B12 deficiency is associated with various clinical conditions including MetS. But its role as independent risk factor for MetS is not clear. Several studies were found association of vitamin B12 with MetS, but result are inconclusive. Based on above information, objective of our study was to evaluate serum vitamin B12 in MetS cases and its association with parameters of MetS.
2. Material and Methods

It was a comparative cross sectional study. This study was conducted at department of biochemistry in collaboration with department of medicine, at Grant Medical College, Mumbai after institutional ethical committee approval. A complete medical history and informed consent were obtained from all participants who were included in the study. Fifty metabolic syndrome cases as per the new IDF criteria of metabolic syndrome in the age group of 30-60 years of either sex and fifty healthy controls in the age groups of 30-60 years of either sex were enrolled in the study. All the cases were taken from general medicine OPD.

2.1. Inclusion criteria

Subjects of both sex and age group between 30 to 60 years were selected in the study. Presence of central obesity (waist circumference ≥ 90 cm in male, ≥ 80 cm in female) and any two of the following factors like decreased HDL level (< 40 mg/dl in males, < 50 mg/dl in females), increased triglyceride level (≥ 150 mg/dl), increased blood pressure (systolic BP ≥ 130 or diastolic BP ≥ 85 mm Hg) and increase FBS level (≥ 100 mg/dl) were selected as cases according to new international diabetes federation (IDF) criteria for MetS.

Exclusion criteria History of previous myocardial infarction, diabetes patients on metformin treatment smoker or tobacco chewer, patients on long term use of proton pump inhibitors, history of hypothyroidism or on its treatment, alcoholics, patient with human immune deficiency virus infection, liver diseases and pregnant women were excluded from the study.

2.2. Withdrawal criteria

Sample collected which became hemolyzed or lipemic, lack of adequate information and insufficient quantity of sample were withdrawn from the study.

After obtaining written consent of the patient, medical history and anthropometric assessment were done. Blood pressure was measured in the sitting position using a mercury sphygmomanometer by auscultatory method. Patients previously diagnosed with and taking medications for hypertension, diabetes or dyslipidemia were also included in the study.

Five ml blood samples were taken from all subjects in morning hour after overnight fasting. Under all aseptic precautions blood was collected by venipuncture in plain vacutainers and fluoride vacutainers. The samples taken in plain vacutainers kept for 30 minutes for clotting after which they were centrifuged at 3000 rpm for 10 minutes to obtain clear serum. Serum sample taken in plain vacutainers were aliquotted in micro centrifuge tubes and stored at -20°C till further assay. The biochemical parameters to be assayed were stable at said temperature for 6-8 weeks. Serum samples were used for the analysis of vitamin B12. Immediate blood glucose estimation was performed from the samples taken in fluoride vacutainers. Estimation of serum vitamin B12, were done on chemiluminescent machine immulite-1000 by using, commercial kits from Siemens Medical Solutions Diagnostics, Los Angeles, CA, USA. Estimation of fasting blood sugar, triglyceride and HDL were done on advia 1800 chemistry autoanalyzer. Waist circumference was measured by measuring tape and blood pressure was measured by sphygmanomaneter.

2.3. Statistical analysis

SPSS version 16 was used for statistical analysis. The graphs and tables were done on ms excel 2010. Pearson’s chi-squared test and Fisher’s exact test were used for quantitative variables. For quantitative variables unpaired-t-test were used when the data were uniformly distributed and mann-whitney U test were used when data were not uniformly distributed. Spearman’s Rho coefficients of correlation were applied in all the correlation, where at least one of the correlate is skewed. P value < 0.05 was considered as a statistical significant.

3. Results

Total hundred subjects were included in study (58 female and 42 male) among which fifty cases (31 female and 19 males) of metabolic syndrome and fifty normal healthy controls (27 female and 23 males) were enrolled in the study. There was no significant difference in gender between cases and controls. (P = 0.418) But 53.44 % of cases were female and 45.23 % cases were male. (Table 1)

The mean age of cases was 48.86 ± 6.28 year and mean age of controls was 45.06 ± 7.01 year. The mean age of case was 3 years more than that of control but this difference of age is not known to produce different biochemical results. (Table 2)

Among cases 38 were Hindus and 12 were Muslim and among controls 40 were Hindus and 10 were Muslim. There was no significant difference in study participant with respect to religion. (Table 3)

The median value of vitamin B12 in cases was 210 pg/ml with the interquartile difference of 293.75 and the median value of vitamin B12 in controls was 178 pg/ml with the interquartile difference of 137.5. The differences between mean ranks of two groups were not significant (Table 4) (Figure 1). Distribution of MetS parameters among cases and controls were described in Table 5.

We further evaluate correlation of vitamin B12 with parameter of MetS. Vitamin B12 were found to be positively correlated with serum triglyceride value (correlation coefficient 0.337; P=0.001) (Figure 2) and serum HDL level. (correlation coefficient 0.207 ; P = 0.039) (Figure 3).
Table 1: Sex wise distribution of metabolic syndrome cases and controls

<table>
<thead>
<tr>
<th>Study group</th>
<th>Sex</th>
<th>Total</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Female</td>
<td>Male</td>
<td></td>
</tr>
<tr>
<td>Cases</td>
<td>31 (62%)</td>
<td>19 (38%)</td>
<td>50(100)</td>
</tr>
<tr>
<td>Controls</td>
<td>27(54%)</td>
<td>23(46%)</td>
<td>50(100)</td>
</tr>
<tr>
<td>Total</td>
<td>58 (58%)</td>
<td>42 (42%)</td>
<td>100(100)</td>
</tr>
</tbody>
</table>

Table 2: Age wise distribution of cases and controls

<table>
<thead>
<tr>
<th>Study group</th>
<th>N</th>
<th>Mean age (Year)</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cases</td>
<td>50</td>
<td>48.86</td>
<td>6.28</td>
</tr>
<tr>
<td>Controls</td>
<td>50</td>
<td>45.06</td>
<td>7.01</td>
</tr>
</tbody>
</table>

Table 3: Religion wise distribution of cases and controls

<table>
<thead>
<tr>
<th>Religion</th>
<th>Cases</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hindu</td>
<td>38 (76%)</td>
<td>40(80%)</td>
</tr>
<tr>
<td>Muslim</td>
<td>12(24%)</td>
<td>10 (20%)</td>
</tr>
<tr>
<td>Total</td>
<td>50 (100%)</td>
<td>50 (100%)</td>
</tr>
</tbody>
</table>

Table 4: Serum Vitamin B12 levels in cases and controls

<table>
<thead>
<tr>
<th>Vitamin B12 (pg/ml)</th>
<th>N</th>
<th>Median</th>
<th>Minimum</th>
<th>Maximum</th>
<th>25th percentiles</th>
<th>75th percentiles</th>
<th>Man-Whitney test P value = 0.25</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cases</td>
<td>50</td>
<td>210</td>
<td>150</td>
<td>1028</td>
<td>159.25</td>
<td>453.00</td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>50</td>
<td>178</td>
<td>151</td>
<td>644</td>
<td>152.00</td>
<td>289.50</td>
<td></td>
</tr>
</tbody>
</table>

Table 5: Distribution of MetS parameters in cases and controls

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Cases (mean ± SD)</th>
<th>Controls (mean ± SD)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Waist circumference (cm)</td>
<td>105.51 ± 7.61</td>
<td>87.26 ± 5.18</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>136.82 ± 18.50</td>
<td>115.56 ± 8.69</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>92.84 ± 10.14</td>
<td>78.28 ± 6.40</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Serum HDL (mg/dl)</td>
<td>26.44 ± 8.25</td>
<td>50.32 ± 6.75</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Serum triglyceride (mg/dl)</td>
<td>184.88 ± 43.85</td>
<td>103.86 ± 35.81</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>FBS (mg/dl)</td>
<td>124.70 ± 42.95</td>
<td>92.96 ± 5.91</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>

There were no significant correlation found with blood sugar level, and blood pressure.

4. Discussion

We conducted study to assess serum vitamin B12 levels in MetS patients and its association with component of the MetS. The median value of serum vitamin B12 in cases and controls were 210 pg/ml and 178 pg/ml respectively. We found that vitamin B12 level was little higher in cases as compared to control but difference was not significant (P=0.25).

It has been found that vitamin B12 treatment in MetS patients improved insulin resistance and endothelial functions. But we excluded cases those were on vitamin B12 treatment in our study. Our findings were similar with Uehara SK et al.(2008), Tungtrongchitr et al (2013) and Reitman et al. (2002) in which there were no statistically significant difference in the vitamin B12 levels in cases with controls.
Pro-inflammatory cytokine like resistin has been found increase in metabolic syndrome and responsible for development of insulin resistance.\textsuperscript{16} It has been reported that resistin level is a predictor of serum vitamin B12 and it was positively correlated with vitamin B12 level.\textsuperscript{17} Slight high level of vitamin B12 in our study might be due to high resistin level in metabolic syndrome.\textsuperscript{17}

We found positive correlation between vitamin B12 and HDL (P=0.039). Disturbance in lipid profile has been found in vitamin B12 deficiency.\textsuperscript{18} Vitamin B12 is a coenzyme for methyl malonyl-CoA mutase enzyme that convert methylmalonyl-CoA to succinyl-CoA. In vitamin B12 deficiency there is an accumulation of methyl malonyl CoA and it is a negative regulator of rate-limiting enzyme of fatty acid oxidation (carnitine palmitoyl transferase) result in increase lipogenesis. High level of homocysteine in vitamin B12 deficiency results in increased level of S-adenosyl L-homocysteine. S-adenosyl L-homocysteine is a negative regulator of S-adenosyl L-methionine dependent methyltransferases and cause hypomethylation of several enzymes and increase level of lipid occurs in muscle and hepatic cell.\textsuperscript{19} These could be the likely mechanism for the link between B12 deficiency and adverse lipid parameters. Our finding supported by Namita Mahalle et al(2013) , they were also found that serum vitamin B12 is positively correlated with HDL levels.\textsuperscript{20,21}

5. Limitations
Study should be done on large population and serum homocysteine and resistin levels should be assessed to validate the results of study.

6. Conclusion
There was no significant difference of serum vitamin B12 levels in metabolic syndrome patients. Se rum vitamin B12 was positively correlated with serum triglyceride and HDL level.

7. Conflict of interest
Authors have no conflict of interest to declare.

8. Funding
This research received no specific funds from any agency.

References


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