Original Research Article

Correlation of Vit B12 levels with metformin usage among type 2 diabetic patients in a tertiary care hospital

Vishnu A. Nareddy, Indeevar P. Boddikuri, Shaik K. Ubedullah*, Abhinaya Papareddy, Manoj Kumar Miriyam

INTRODUCTION

T2DM is a metabolic disorder that is increasingly becoming a public health concern. The disease is associated with a variety of systemic macrovascular and microvascular complications. Diabetic peripheral neuropathy (DPN) is the most common complication, and it may eventually develop in up to 50% of patients. It is assumed that prolonged use of metformin causes vitamin B12 deficiency. The clinical presentation of vitamin B12 deficiency generally includes haematological and neurological manifestations. The aim of the study is to correlate vit b12 levels in relation to duration of usage of metformin for a period of more than 1 year.

ABSTRACT

Background: T2DM is a metabolic disorder that is increasingly becoming a public health concern. The disease is associated with a variety of systemic macrovascular and microvascular complications. Diabetic peripheral neuropathy (DPN) is the most common complication, and it may eventually develop in up to 50% of patients. It is assumed that prolonged use of metformin causes vitamin B12 deficiency. The clinical presentation of vitamin B12 deficiency generally includes haematological and neurological manifestations. The aim of the study is to correlate vit b12 levels in relation to duration of usage of metformin for a period of more than 1 year.

Methods: The study was conducted from November 2017 to June 2018 for a period of 7 months which included 50 subjects from both sex groups, aged 18 years and above, on metformin with subjective evidence of neuropathy and compared with vit b12 levels.

Results: The study results indicate that there is no significant drop in vit b12 levels in metformin users in significant proportions to cause vitb12 deficient-neuropathy. The p value being 0.658 comparing vit b12 with metformin usage years (<2 yrs, 2-5yrs, 5-8yrs, ≥8yrs).

Conclusions: This cross-sectional study indicates that diabetic patients who were on metformin were not having any vit b12 deficiency, though all patients had subjective neuropathy. This suggests that neuropathy is not related to metformin induced Vit B12 deficiency. Therefore, prescribing of vit b12 supplementation in any long term metformin treated patients is no longer necessary and reduces patients financial burden.

Keywords: Metformin, Peripheral neuropathy, Type 2 diabetes mellitus, Vitamin B12
confirmed this association.\textsuperscript{7-11} Since vitamin B12 is essential for the remethylation of homocysteine to methionine, metformin-induced vitamin B12 deficiency could be associated with hyperhomocysteinemia, a condition with a questionable detrimental impact on macrovascular disease in T2DM patients.\textsuperscript{12}

Metformin acts primarily at the liver by reducing glucose output and, secondarily, by augmenting glucose uptake in the peripheral tissues, chiefly muscle. These effects are mediated by the activation of an upstream kinase, liver kinase B1 (LKB-1), which in turn regulates the downstream kinase adenosine monophosphatase protein kinase (AMPK). AMPK phosphorylates a transcriptional co-activator, transducer of regulated CREB protein 2 (TORC2), resulting in its inactivation which consequently downregulates transcriptional events that promote synthesis of gluconeogenic enzymes.\textsuperscript{13} Inhibition of mitochondrial respiration has also been proposed to contribute to the reduction of gluconeogenesis since it reduces the energy supply required for this process.\textsuperscript{14} The clinical presentation of vitamin B12 deficiency generally includes haematological and neurological manifestations. Neuropathy can be the only manifestation of the deficiency, without a haematologic presentation.\textsuperscript{15} It is also well known that Vitamin B12 deficiency also causes hematological anemia.\textsuperscript{16} It is however noteworthy that subtle neurological manifestation appears earlier than hematological changes.\textsuperscript{17}

India has more people with diabetes than any other country in the world.\textsuperscript{18} The pre-existing cobalamine deficiency as a result of dietary habits poses an additional burden of neuropathy.\textsuperscript{19} Animal studies have recently shown that metformin can exert neuroprotective and antineuropathic activities that are independent of its euglycemic effect. Metformin protects against numbness and neuropathic pain induced by chemotherapy in mice.\textsuperscript{20} The sensory symptoms of pain, dyasesthesia, and paraesthesia characterize both diabetic PN and chemotherapy-induced PN, signifying the impact of these findings. Animal studies have also reported that metformin abolished pain resulting from the activation of sensory neurons as well as resolved neuropathic allodynia, protected against ethanol-induced neuronal apoptosis, and enhanced neurogenesis.\textsuperscript{21-24} It also suppressed cortical neuronal apoptosis and exerted neuroprotective effects in Parkinson’s disease.\textsuperscript{25,26}

In this study we are aiming to compare vitamin B12 levels in T2DM patients and duration of Metformin usage and also its correlation in different age groups and both sexes.

**METHODS**

This Prospective hospital-based study extended from November 2017 to June 2018. A total of 50 patients were studied who were attending Medicine OPD and inpatients in the medicine department of Narayana Medical College, Nellore, Andhra Pradesh, India. Vit B12 levels were collected. Complete hemogram was also done to exclude pernicious anaemia and any cellular level changes in blood cells due to borderline Vit B12 deficiency. Normal Vit B12 levels range from 200-832, of which 200-250 were considered borderline, and below 200 were considered deficient.

The patients included in the study group fulfilled the following inclusion criteria:

- Type 2 DM patients, 18 years and above who fulfilled WHO diagnostic criteria (Blood Sugar Fasting >126 or Blood Sugar >200, 2 h after glucose challenge) and
- Patients who were on metformin treatment for at least one year.

The exclusion criteria included patients who were non-consenting; had type 1 diabetes mellitus; had other causes of cobalmine deficiency like malabsorption syndrome or Gastro-Intestinal surgery, Pernicious anaemia, chronic kidney disease and who are “at risk” and “heavy” alcoholics according NIAAA (National Institute on Alcohol Abuse and Alcoholism) Criteria. Approval for this study was received from the ethics board.

**RESULTS**

Patients using metformin were grouped under 4 categories of <2 years, 2-5 years, 5-8 years, ≥8 years and the mean Vit B12 levels in each category are 627.25, 496.87, 394.44, 498.60 respectively (Table 1) with normal range being 200-832 and the total mean of all the groups being 475.04 which indicates there has been no significance of metformin on Vit B12. P value 0.658 that is not significant (Table 1).

![Table 1: Comparison of Vit B12 levels among the metformin groups.](image-url)

**Table 1: Comparison of Vit B12 levels among the metformin groups.**

<table>
<thead>
<tr>
<th>Grouping of patients on metformin</th>
<th>No. of patients</th>
<th>Mean vitamin B12 level</th>
<th>Std. deviation</th>
<th>F value</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;2 Years</td>
<td>4</td>
<td>627.25</td>
<td>277.25</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2-5 Years</td>
<td>15</td>
<td>496.87</td>
<td>385.598</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5-8 Years</td>
<td>16</td>
<td>394.44</td>
<td>321.114</td>
<td>0.540</td>
<td>0.658 (Not Sig.)</td>
</tr>
<tr>
<td>≥8 Years</td>
<td>15</td>
<td>498.60</td>
<td>393.468</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>50</td>
<td>475.04</td>
<td>356.813</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
When depicted graphically there has been a substantial fall in Vit B12 levels among the patients in 5-8 years of metformin usage category, when compared with <2 years, 2-5 years. There was a peak initially and progressive fall is noted when it came to 2-5 year category (Figure 1).

![Figure 1: Vit B12 levels with metformin years.](image)

Present study includes total of 50 patients with males=34, females=16 patients, when analysed in both males and females. Of all the categories, 5-8 years category show decreased level of Vit B12 of mean 404.69 in males and 350.00 in females. Mean vitamin B12 levels in males and females are 478.00 and 468.75 respectively (Table 2).

<table>
<thead>
<tr>
<th>Grouping of patients on metformin in years</th>
<th>Sex</th>
<th>Mean vitamin B12 levels</th>
<th>Std. deviation of Vit B12</th>
<th>No. of patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;2 Years</td>
<td>Males</td>
<td>542.30</td>
<td>467.101</td>
<td>10</td>
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<td>2-5 Years</td>
<td>Males</td>
<td>404.69</td>
<td>349.754</td>
<td>13</td>
</tr>
<tr>
<td>≥8 Years</td>
<td>Males</td>
<td>506.18</td>
<td>453.303</td>
<td>11</td>
</tr>
<tr>
<td>Total</td>
<td>Males</td>
<td>478.00</td>
<td>412.177</td>
<td>34</td>
</tr>
<tr>
<td>&lt;2 Years</td>
<td>Females</td>
<td>627.25</td>
<td>277.257</td>
<td>4</td>
</tr>
<tr>
<td>2-5 Years</td>
<td>Females</td>
<td>466.00</td>
<td>118.341</td>
<td>5</td>
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<tr>
<td>≥8 Years</td>
<td>Females</td>
<td>350.00</td>
<td>189.058</td>
<td>3</td>
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<tr>
<td>Total</td>
<td>Females</td>
<td>477.75</td>
<td>191.681</td>
<td>4</td>
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<td>&lt;2 Years</td>
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<tr>
<td></td>
<td>Total</td>
<td>475.04</td>
<td>356.813</td>
<td>50</td>
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</table>

DISCUSSION

The 2018 ADA Clinical Practice Recommendations endorse screening Metformin users for vitamin B12 deficiency and the 2017 ADA diabetic neuropathy statement recommends that all patients with diabetic neuropathy should be assessed for B12 deficiency, to exclude a treatable cause of neuropathy. Lower prevalence of B12 deficiency in patients taking Metformin, were assessed confounding factors such as other medications, which may alter B12 levels. Sulfonylurea use in combination with Metformin is a significant independent risk factor for B12 deficiency.

Vitamin B12 deficiency can lead to clinically significant but treatable conditions. These include a wide range of clinical conditions, such as memory impairment, peripheral neuropathy, dementia, delirium, sub acute combined degeneration of the spinal cord, megaloblastic anemia and pancytopenia. Accumulating evidence suggests that long-term use of metformin is associated with low vitamin B12 levels, and findings from both observational and interventional studies have confirmed this association.

As far as our study is concerned there has been no correlation of metformin with Vit B12 levels which maybe supporting as well as contradicting some studies. in our study no. of patients on metformin for >8 years in both males and females are 11 (22%) and 4 (8%) constituting a total of 30% of population. further studies should be encouraged to conduct vit B12 levels in prolonged metformin users of more than 10 years to substantiate any significant correlation.

Clinically, a retrospective chart review also reported the association between metformin use and a decrease in lumbar radiculopathy pain. Antineuropathic effects of metformin may be mediated by 5-adenosine monophosphate-activated protein kinase (AMPK) activation.

Impaired AMPK signaling was linked to PN in animals. So, in contrary to the above mentioned citations and research, there has been exerted a neuroprotective effect in animal studies, and not yet confirmed on humans. If there is neuroprotectic effect in metformin users, molecular level evidence is not yet found to substantiate. Whereas animal studies 5AMPK signaling impairment was found to be the culprit.

However, unless proven otherwise vitamin b12 supplementation in metformin users must be withheld causing a financial burden among many patients. Many patients suffering from pernicious anemia with type 2 diabetes mellitus have been with held metformin even it is first line of treatment according to current guidelines.

Limitations of this study were that previous vitamin supplementation history was not available among patients and neuropathy in patients was completely subjective and no objective evidence is available.
CONCLUSION

There has been a negative correlation in between vitamin B12 levels and metformin usage. There was no significant reduction in the patients even on prolonged usage of 5-8years. In the metformin usage patients for more than 8 years have a normal levels when compared to previous group. Thereby indicating that our study shows there is no significance of vitamin B12 supplementation in metformin users as these are not the culprits in occurrence of peripheral neuropathy. Financial burden both for patients and government can be reduced to a great extent since it takes a great amount of effort and money to supply a broad base of patients for a long-term of period.

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Conflict of interest: None declared
Ethical approval: The study was approved by the Institutional Ethics Committee

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