Original Research Article

Prevalence of vitamin B12 deficiency in Indian type 2 diabetes subjects on metformin therapy

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Abstract

Background: To find out the prevalence of vitamin B12 deficiency in Indian type 2 diabetes subjects on metformin therapy.**Material & Methods**: 161 type 2 diabetes subjects were studied over a period of 6 months at Karnataka institute ofendocrinology Bangalore. All subjects gave written informed consent. BMI, Waist circumference, FPG, PPPG and HBA1c were estimated. Vitamin B12 levels were estimated by electrochemiluiminescence. We have excluded patients taking alcohol, vitamin B12 supplements, pregnant woman, and type 1 diabetics. Subjects whose vitamin B12 less than 200 picogram/ml were considered to be deficient in vitamin B12. Subjects who were on metformin treatment for more than 6 months were included in the study.**Results:**118 diabetes subjects were males. They were in the age group of 30 to 80 years. Duration of diabetes was 1 to <5 years in 20.5%, 5 to <10 years in 28.5% and 10 years and morein 51% of diabetes subjects studied respectively. 55.9% of subjects had positive family history of type 2 diabetes. 112 subjects were on 1000 mg metformin for one year or more and 49 were on 2000 mg metformin for one year or more. Prevalence of vitamin B12 deficiency was 27.33%.**Conclusions:** The prevalence of vitamin B12 deficiency is 27.33% in type 2 diabetes subjects on metformin therapy. The percentage of deficiency increased with increase in dosage of metformin but there was no correlation to duration of metformin therapy.

Keywords: vitamin B12, Metformin, Type 2 diabetes

Introduction

Vitamin B12 or cobalamin is a water-soluble vitamin that plays a very fundamental role in DNA synthesis, optimal haemopoesis and neurological function. The clinical picture of vitamin B12 deficiency hence, is predominantly of features of haematological and neuro-cognitive dysfunction [1].

The proposed mechanisms to explain metformin induced vitamin B12 deficiency among patients with T2DM include: alterations in small bowel motility which stimulates bacterial overgrowth and consequential vitamin B12 deficiency, competitive inhibition or inactivation of vitamin B12 absorption, alterations in intrinsic factor (IF) levels and interaction with the cubulin endocytic receptor[2]. Metformin has also been shown to inhibit the calcium dependent absorption of the vitamin B12-IF complex at the terminal ileum. This inhibitory effect is reversed with calcium supplementation [3]. Decrease in vitamin B12 absorption and levels following metformin use typically starts as early as the 4th month [4]. There is a large storage of vitamin B12 in the liver so overt clinical features manifest by 5 to 10 years [2].

Screening approach for vitamin B12 deficiency among patients with T2DM- Currently, there are no published guidelines advocating for routine screening for vitamin B12 deficiency among patients with T2DM.

Manuscript received: 6th September 2017
Reviewed: 15th September 2017
Author Corrected: 24th September 2017
Accepted for Publication: 30th September 2017

However, among type 2 diabetic patients, it is clinically plausible to screen for vitamin B12 deficiency prior to initiation of metformin and later annually among elderly patients with history of long term use of metformin ≥3-4 years, use of high doses of metformin (≥2 g/day), clinically worsening diabetic distal polyneuropathy in the presence or absence of the haematological abnormalities [5].

The screening approach for vitamin B12 deficiency among diabetic patients and the general population is similar. Measurement of the serum vitamin B12 concentrations should be the preliminary screening step for vitamin B12 deficiency among patients with T2DM. Concentrations <200 pg/ml are usually diagnostic of vitamin B12 deficiency while concentrations >400 pg/ml confirm absence of vitamin B12 deficiency [6].

Measurement of serum MMA or homocysteine concentrations is a more sensitive and specific approach for screening especially among type 2 diabetic patients with borderline serum vitamin B12 concentrations of 200-400 pg/ml and subtle haematological manifestations.

Serum homocysteine and methylmalonic acid concentrations of 5-15 µmol/l and <0.28 µmol/l are considered within the normal range respectively[5,7].

Reinstatler et al. in the National Health and Nutrition Examination Survey of 1999–2006 in the USA defined Biochemical B_{12} deficiencyas serum levels ≤148 pmol/L, borderline deficiency as serum B_{12} >148 to ≤221 pmol/L, and normal as >221 pmol/L(400 Pmol/L=550pg/ml [8].

Research methods-Study design-161 type 2 diabetes subjects were studied over a period of 6 months at Karnataka institute of endocrinology and research Bangalore.All subjects gave written informed consent. BMI, Waist circumference, FPG, PPPG and HBA1c were estimated. Vitamin B12 levels were estimated by electrochemiluiminescence.

Inclusion criteria-Patients with type 2 diabetes, aged 30 to 80 yr, who had taken metformin for at least six months were recruited at Karnataka institute of endocrinology.

Exclusion criteria included patients with newly diagnosed type 2 diabetes, patients who had pernicious anemia, pregnant women, type 1 diabetes, decreased renal function (serum creatinine levels > 1.5 mg/dL for men and > 1.4 mg/dL for women), gastrectomy, colectomy, inflammatory bowel disease, Patients were also excluded if they had any severe medical illness, such as sepsis, severe infection, malignancy, liver cirrhosis, heart failure, or renal failure patients taking alcohol, vitamin B12 supplements.

Statistical methods**-** Descriptive and inferential statistical analysis has been carried out in the present study. Results on continuous measurements are presented on Mean ± SD (Min-Max) and results on categorical measurements are presented in Number (%). Significance is assessed at 5% level of significance.

The following assumptions on data is made, **Assumptions:**

1. Dependent variables should be normally distributed.

2. Samples drawn from the population should be random, Cases of the samples should be independent Chi-square/ Fisher Exact test has been used to find the significance of study parameters on categorical scale between two or more groups.

In this study we have used values of

<200 pg/m/ml fordefinite vitamin B12 deficiency
>200 to 300 pg/ml for borderline vitamin B12 deficiency.
>300 pg/ml for normalvitamin B12 levels

Results-118 diabetes subjects were males. They were in the age group of 30 to 80 years. Table 1

Table- 1: Age distribution of patients studied.

Age in years	Gender		Total
	Female	Male	
<40	1(2.3%)	3(2.5%)	4(2.5%)
40-50	9(20.9%)	27(22.9%)	36(22.4%)
51-60	21(48.8%)	37(31.4%)	58(36%)
61-70	12(27.9%)	39(33.1%)	51(31.7%)
71-80	0(0%)	12(10.2%)	12(7.5%)
Total	**43(100%)**	**118(100%)**	**161(100%)**

BMI was less than 25 in 41% of patients, 25 to 30 in 44.7% and more than 30 in 14.3% of diabetes patients. Table 2

Table- 2: BMI (kg/m^2) distribution of patients studied.

BMI (kg/m^2)	Gender		Total
	Female	Male	
<18.5	0(0%)	0(0%)	0(0%)
18.5-25	7(16.3%)	59(50%)	66(41%)
25-30	23(53.5%)	49(41.5%)	72(44.7%)
>30	13(30.2%)	10(8.5%)	23(14.3%)
Total	**43(100%)**	**118(100%)**	**161(100%)**

Duration of diabetes was 1 to <5 years in 26.1%, 5 to <10 years in 34.8% and 10 years and morein 39.1% of diabetes subjects studied respectively. 55.9% of subjects had positive family history of type 2 diabetes.

Table- 3: Vitamin B12 of patients studied.

Vitamin B12	Gender		Total
	Female	Male	
<200	9(20.9%)	35(29.7%)	44(27.3%)
200-300	9(20.9%)	34(28.8%)	43(26.7%)
>300	25(58.1%)	49(41.5%)	74(46%)
Total	**43(100%)**	**118(100%)**	**161(100%)**

Table- 4: Metformin dose of patients studied.

Metformindose	Gender		Total
	Female	Male	
1000	34(79.1%)	78(66.1%)	112(69.6%)
2000	9(20.9%)	40(33.9%)	49(30.4%)
Total	**43(100%)**	**118(100%)**	**161(100%)**

Table- 5: Vitamin B12 in relation to Metformin duration.

Vitamin B12	Metfor min duration			Total
	<5	5-10	>10	
<200	27(33.3%)	10(21.3%)	7(21.2%)	44(27.3%)
200-300	20(24.7%)	13(27.7%)	10(30.3%)	43(26.7%)
>300	34(42%)	24(51.1%)	16(48.5%)	74(46%)
Total	**81(100%)**	**47(100%)**	**33(100%)**	**161(100%)**

112 subjects were on 1000 mg metformin for one year or more and 49 were on 2000 mg metformin for one year or more. Prevalence of vitamin B12 deficiency was 27.33%. Subgroup analysis showed that 23.2% on 1000 mg metformin and 36.73% on 2000 mg metformin were deficient in vitamin B12 respectively. There was no correlation between vitamin B12 deficiency and duration of metformin therapy. Table 3,4,5.

Discussion

According to ADA-EASD consensus, AACE, IDF and NICE guidelines metformin is the first drug of choice in type 2 diabetes unless contraindicated or not tolerated.Several cross-sectional studies [9,10] and case reports [11,12] have documented an increased frequency of vitamin B12 deficiency among type 2 DMpatients. Metformin use has been unequivocally demonstrated as the prime factor associated with vitamin B12 deficiency among patients with T2DM [13]. Studies assessing type 2 diabetic patients on metformin have reported the prevalence of vitamin B12 deficiency to range from 5.8% to 33% [8,13,14].

This wide variation in the reported prevalence could probably be explained by the varied study definitions of vitamin B12 deficiency. In the cross sectional study by Pflipsen et al. on 203 outpatient type 2 diabetic patients at a large military primary care clinic in USA, definite vitamin B12 deficiency was defined as serum vitamin B12 concentrations of <100 pg/ml or elevated serum methylmalonic acid of >243 nmol/L or homocysteine concentrations of >11.9 nmol/L if serum vitamin B12 concentrations were between 100 to 350 pg/mL [8]. In one cross sectional study that documented a high prevalence of vitamin B12 deficiency of 33% among adult patients with T2DM by Qureshi et al., vitamin B12 deficiency was defined as serum vitamin B12 concentrations <150 pg/ml [14], However, patients enrolled in this study were those who were on high dose (>2 g/day) and long-term (4 years) metformin treatment, both clinical factors known to be associated with vitamin B12 deficiency.

Due to the diverse definitions of vitamin B12 deficiency used in most studies and the cultural and religious beliefs in different regions of the world, comparison of the prevalence of vitamin B12 deficiency among T2DM patients and healthy general populations is difficult.

In India, a country with a large proportion of vegetarians due to cultural and religious beliefs, very high prevalence of vitamin B12 deficiency among the general population has been reported. In one study by Yajnik et al. to determine the frequency of vitamin B12 deficiency and hyper homo cysteinemia among 441 healthy middle aged Indian men, vitamin B12 deficiency as defined by vitamin B12 concentrations <150 pmol/L was reported among 67% of the study participants [15]. In another cross sectional study among 175 healthy elderly Indian subjects aged >60 years, vitamin B12 deficiency also defined as vitamin B12 concentrations <150 pmol/L was reported among 16% of the study participants [16]. In one early randomised controlled trial by DeFronzo et al., metformin decreased the serum vitamin B12 levels by 22% and 29% compared to placebo and glyburide respectively [17]. A recent, randomized control trial designed to examine the temporal relationship between metformin and serum B_{12} found a 19% reduction in serum B_{12} levels compared with placebo after 4 years [18]. Although classical B_{12} deficiency presents with clinical symptoms such as anaemia, peripheral neuropathy, depression, and cognitive impairment, these symptoms are usually absent in those with biochemical B_{12} deficiency[19].

Vitamin B12 deficiency is clinically important because it is a reversible cause of bone marrow failure and demyelinating nerve disease. Neurologic damage, a possible consequence of metformin-induced vitamin B12 deficiency, can present as peripheral neuropathy and may be mistaken for diabetic neuropathy in patients on metformin treatment [20].

Low vitamin B12 levels have been reported to be associated with worse nerve conduction velocities and poorer responses to light touch by monofilament detection [21] This may lead to the unnecessary use of anticonvulsants or tricyclic antidepressants[20,22.23]. Another study explored the relationship between low serum vitamin B12 levels and cognitive impairment, depression and neuropathy. Low vitamin B12 states were more associated with symptoms of memory impairment with objective evidence of cognitive impairment

than with depression or neuropathy[24]. Prevalence of definite vitamin B12 deficiency in 27.3% andbiochemical B12 deficiency in 26.3% is seen in the present study. Subgroup analysis showed that 23.2% on 1000 mg metformin and 36.73% on 2000 mg metformin were deficient in vitamin B12 respectively.

There was no correlation between vitamin B12 deficiency and duration of metformin therapy. As vitamin B12-associated neuropathy is a treatable and reversible condition, early detection and treatment of vitamin B12 deficiency is clinically important in patients with diabetes using metformin.

Conclusions

The prevalence of vitamin B12 deficiency is 27.33% in type 2 diabetes subjects on metformin therapy. The percentage of deficiency increased with increase in dosage of metformin but there was no correlation to duration of metformin therapy. Clinically if the physicians suspect vitamin B12 deficiency in type 2 diabetes subjects on metformin therapy he should do vitamin B12 assay and accordingly treat with oral or injection vitamin B12.

Acknowledgements- Dr. K. P. Suresh, Scientist (Biostatistics), National Institute of Animal Nutrition & Physiology, Bangalore-560030. No potential conflicts of interest relevant to the article are reported.

Funding: Nil, Conflict of interest: None
Permission of IRB: Yes

Abbreviations

BMI- Body mass index.Type2 DM- Type 2 diabetes mellitus. ADA-American diabetes association. EASD-European association of study of Diabetes. AACE-American association of college of endocrinologyIDF-International diabetes federation

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How to cite this article?

Anil Kumar R, Surekha B. Shetty, Lalitha R. Prevalence of vitamin B12 deficiency in Indian type 2 diabetes subjects on metformin therapy. *Int J Med Res Rev* 2017;5(09):845-850.doi:10.17511/ijmrr. 2017.i09.03.

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