RESEARCH ARTICLE

Vitamin B12 status and peripheral neuropathy in patients with type 2 diabetes mellitus

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Abstract

Objective: To assess the status of vitamin B12 in patients with type 2 diabetes, and to explore any association between its deficiency and diabetic peripheral neuropathy.

Methods: This cross-sectional observational study was conducted from August, 2017, to April, 2018, at the Specialized Centre for Endocrinology and Diabetes in Baghdad, Iraq. Type 2 diabetics using metformin were subjected to clinical examination for retinopathy using fundoscopy, and peripheral neuropathy using the Michigan Neuropathy Screening Instrument. Additionally, patients were asked to fill a questionnaire and their medical records were reviewed. Blood samples were obtained for the measurement of biomarkers. Vitamin B_{12} deficiency was recorded at ≤187 pg/ml. Data was analysed using SPSS 25.

Results: Of the 66 patients, 39(59%) were males and 27(41%) were females. The overall mean age was 53.3±9.2 years and the mean duration of diabetes was 104±71.8 months. The mean dose of metformin was 1135±496 mg and the duration of metformin use was 72±62 months. Overall, 19(29%) patients suffered from vitamin B_{12} deficiency. However, no significant difference was found between normal and deficit groups regarding the parameters that may affect vitamin B_{12} level. Also, no significant correlations were found between vitamin B_{12} concentration and the dose (p=0.16) or the duration of metformin use (p=0.09).

Conclusions: High prevalence of vitamin B_{12} deficiency was observed in metformin-treated patients with type 2 diabetes. However, the deficiency had no correlation with the rate of peripheral neuropathy.

Keywords: Vitamin B_{12}, Metformin, Type 2 diabetes. (JPMA 69: S-40 (Suppl. 3); 2019)

Introduction

Type 2 diabetes mellitus (T2DM) is regarded as one of the most common non-communicable diseases all over the world. In 2015, the International Diabetes Federation (IDF) estimated that 415 million people worldwide had diabetes. The number of people with diabetes is expected to reach 642 million by 2040;an increase of 55%.[1,2]

Diabetic polyneuropathy is primarily a symmetrical sensory polyneuropathy, initially affecting the distal lower extremities. There is evidence of nerve damage at the time of diagnosis of diabetes in 10-18% of patients, suggesting that even pre-diabetes is associated with neuropathy.[3]

In the absence of contraindications, metformin is considered the first-line T2DM treatment according to the 2006 Consensus Statement from the American Diabetes Association (ADA) and the European Association for the Study of Diabetes (EASD).[4] Metformin is effective only in the presence of insulin, and its major effect is to decrease hepatic glucose synthesis, also called gluconeogenesis.[5,6]

Vitamin B_{12} (cobalamin, Cb1) is a water-soluble vitamin principally obtained from animal proteins like meat and dairy products.[7] The usual Western diet contains 5-7mcg of Cb1per day, which is enough to maintain normal Cbl equilibrium.[8] For adults, the recommended dietary allowance (RDA) for Cbl is 2mcg/day. Vitamin B_{12} is required for deoxyribonucleic acid (DNA) synthesis, red blood cell (RBC) production, and neurological function.[9]

Metformin reduces intestinal absorption of vitamin B_{12} in up to 30% patients, and lowers serum vitamin B_{12} concentrations in 5-10%, but only rarely causes megaloblastic anaemia.[10] Peripheral neuropathy may precede the development of megaloblastic anaemia in some patients with vitamin B_{12} deficiency.[11] Studies describing the link between vitamin B_{12} deficiency and peripheral neuropathy in patients with T2DM are lacking and have produced conflicting results.[12,13]

The current study was planned to determine the prevalence of vitamin B_{12} deficiency, and to ascertain whether this deficiency affects the incidence of peripheral neuropathy in T2DM patients.

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Patients and Methods

Cross-sectional observational study was conducted from August, 2017, to April, 2018, at the Specialized Centre for Endocrinology and Diabetes in Baghdad, Iraq. After approval from the Iraqi Scientific Council of Internal Medicine, T2DM patients on metformin for >6 months were enrolled. Those excluded were patients with estimated glomerular filtration rate (eGFR) <60ml/min/1.73m^2, intake of multivitamin or vitamin B_{12}, acid-lowering agents such as proton pump inhibitors (PPIs) or H2 receptor antagonist, as well as alcoholics and pregnant women in addition to patients with co-morbidities, such as liver disease, psoriasis, tumours and leukaemia, pernicious anaemia, human immunodeficiency virus (HIV), mal-absorptive conditions, like coeliac disease, inflammatory bowel disease, gastrointestinal surgery, including gastrectomy and gastric bypass surgery, and malnutrition owing to pure vegans, pancreatic insufficiency and anorexia nervosa.

After verbal consent form the subjects, demographic and clinical data was recorded. This included age, gender, duration of diabetes, medication and smoking history, cardiovascular and renal co-morbidities and diabetic foot and eye surgery history. Anthropometric data were obtained by measuring height, weight, waist circumference (WC) and body mass index (BMI). Thorough physical examination included measurements of blood pressure (BP) by standardised aneroid sphygmomanometer and fundoscopic eye examination (Keller ophthalmoscope) for retinopathy which was done by two researchers to confirm the findings.

Peripheral neuropathy was diagnosed according to the Michigan Neuropathy Screening Instrument,[14] which is a tool for the diagnosis of diabetic neuropathy in outpatient clinics.[15] The test looks for certain information and grade them on a point-based system[14,15] as follows:

Do the feet show dry skin, callus, fissure, infection or deformities? The presence of any of these indicators of neuropathy is scored as 1 point and an additional point is added if an ulcer is present.

What is the vibration sense on the dorsum of the great toes? Reduced = 0.5 point; absent = 1 point.

What is the Achilles tendon reflex? Absent = 1 point; present with reinforcement = 0.5 points.

This test was standardized against the San Antonio Consensus criteria. A score >2 indicated neuropathy with 95% specificity and 80% sensitivity.[16]

Blood samples were obtained from patients to measure the following biomarkers: vitamin B_{12} (Architect i2000sr; Abbott Laboratories, Abbott Park, IL, USA), fasting blood glucose (FBG), glycated haemoglobin (HbA1C), creatinine, lipid profile, haemoglobin (Hb) (Cobas C311; Roche Diagnostics, Germany), and mean corpuscular volume (MCV) (ABX Micros ES -60 haematology analyser, Japan). The cut-off value of vitamin B_{12} was lower than or equal to 187 pg/ml.

Data was analysed using SPSS 25. Continousvariables were presented as mean ± standard deviation (SD). Chi square test were used for discrete variables and was expressed as frequencies and percentages. P<0.05 was considered statistically significant.

Results

Of the 66 patients, 39(59%) were males and 27(41%) were females. The overall mean age was 53.3±9.2 years and the

Table-1: Demographic and clinical characteristics of the participants.

Demographic Characteristic	Value*
Age (years)	53.3±9.2
Gender	
Male	27(40.9%)
Female	39(59.09%)
Duration of DM(months)	104±71.8
Dose of metformin (mg/d)	1135.6±496.6
Duration of metformin use(months)	71.8±62.1
Height(cm)	161.9±9.8
Weight(kg)	77.7±14
BMI(kg/m^2)	29.8±6
Waist circumference (cm)	102.5±10.8
SBP(mmHg)	142.5±29
DBP(mmHg)	85±13.8
eGFR(ml/min/1.73m^2)	100±14
Creatinine(mg/dl)	0.7±0.1
FBG(mg/dl)	220±81
HbA1c %	9.8±1.8
LDL(mg/dl)	99.5±44.4
HDL(mg/dl)	41.9±11.5
TG(mg/dl)	253±255.8
MCV(FL)	83.6±7.4
Hb(g/dl)	13.3±1.4
Smoking	11 (16.6%)
Cardiovascular diseases	13 (59.09%)
Hypertension	33 (50%)

*Data presented either as mean±SD or number (percentage).
BMI: Body mass index
SBP: Systolic blood pressure
DBP: Dystolic blood pressure
eGFR: Estimated glomerular filtration rate
FBG: Fasting blood glucose
HbA1c: Glycated haemoglobin
LDL: Low-density lipoprotein
HDL: High-density lipoprotein
TG: Triglycerides. MCV: Mean corpuscular volume. Hb: Haemoglobin.

Table-2: Comparison of the participants' characteristics according to their vitamin B12 status*.

Variable	B12 Status		P value**
	Normal	Deficit	
Gender			
Female	28	11	
Male	19	8	0.9
Hypertension			
Yes	25	8	
No	22	11	0.4
Smoking			
Yes	7	4	
No	40	15	0.7
Insulin			
Yes	7	4	
No	40	15	0.7
Cardiovascular disease			
Yes	9	4	
No	38	15	1
BMI (kg/m^2)			
<28	21	10	
≥28	26	9	0.5
Duration of diabetes (months)			
<96	20	12	
≥96	27	7	0.1
Duration of metformin (months)			
≤12	7	3	
>12	40	16	1
Dose of metformin (mg/day)			
≤1000	32	11	
>1000	15	8	0.4
Retinopathy			
Yes	6	1	
No	41	18	0.6
Hb (g/dl)			
<13	15	6	
≥13	31	13	0.9
MCV (FL)			
<85	25	5	
≥85	21	12	0.09

*BMI: Body mass index; Hb: Haemoglobin;MCV: Mean corpuscular volume.
**Differences between groups measured using Chi Square or Fisher exact test.

mean duration of diabetes was 104±71.8 months. The mean dose of metformin was 1135±496 mg and the duration of metformin use was 72±62 months (Table-1). Cardiovascular diseases were reported in 13(17%) patients while 33(50%) were hypertensive. Peripheral neuropathy was detected in 30(46%) patients, and 7(11%) showed only signs of retinopathy.

Mean vitamin B_{12} concentration was 318.3±184.3 pg/ml and 19(29%) patients had vitamin B_{12} deficiency, but there was no significant difference between normal and deficit groups regarding the parameters that may affect vitamin B_{12} level (Table-2). Also, no significant correlations were found between vitamin B_{12} concentration and the dose (beta: 0.06, p=0.16) or the duration of metformin use (beta: 0.61, p=0.09).

However, those with neuropathy had significantly longer duration of diabetes (p= 0.04), higher systolic blood pressure (SBP) (p=0.05) and lower serum Hb concentrations (p=0.03).

Discussion

The study observed vitamin B_{12} deficiency in 29% participants and 30(46%) were suffering from peripheral neuropathy. However, no correlation was found between vitamin B_{12} concentration and the incidence of peripheral neuropathy (p>0.05).

Also, there was no statistically significant association between the dose or the duration of metformin intake and vitamin B_{12} concentration. Earlier studies also reported no significant association between the concentration of vitamin B_{12} and the dose or the duration of intake of metformin.[17,18] In contrast, several other studies reported significant correlation between metformin dose or duration of use and vitamin B_{12} concentration.[10,19-21] This variation in the results might be related to the various study designs and populations included in these trials. Some investigators did not follow strict exclusion criteria by including the elderly and those using PPIs or H2 antagonists. This may confound the results regarding the effects of metformin on vitamin B_{12} concentration.[17]

A few previous studies have demonstrated that the decrease in serum B_{12} levels occurs within 3-4 months after the commencement of metformin treatment.[22] However, according to most reports, vitamin B_{12} deficiency occurs only after 5-10 years of metformin usage.[13] This delay in the appearance of B_{12} deficiency may be due to the significant hepatic stores of this vitamin.[23] In line with a study, the current study did not find significant correlation between vitamin B_{12} concentration and the duration of diabetes.[17]

In the current study, there was no correlation of vitamin B_{12} deficiency with the incidence of peripheral diabetic neuropathy which is in line with earlier findings.[24] However, in the current study, the absence of correlation of peripheral diabetic neuropathy with vitamin B_{12} deficiency should be interpreted with caution as no objective procedures were undertaken to diagnose neuropathy.

Two case reports have shown metformin-related B_{12} deficiency presenting as peripheral neuropathy.[11]

According to such reports, it has been suggested that annual vitamin B_{12} supplementation should be given to patients receiving metformin therapy, and this has become part of recognised empirical practice.[25]

Anaemia and MCV are well-known clinical parameters indicative of vitamin B_{12} deficiency. A case of megaloblastic anaemia secondary to vitamin B_{12} mal-absorption and long-term metformin treatment has been reported. Anaemia tends to occur only when the deficiency is severe enough to affect the haematological indices. In addition, macrocytosis can be masked by coexisting microcytic processes, including thalassemia and iron deficiency.[26]

The current study has some limitations. First, the dietary intake of vitamin B_{12} was not recorded. However, information usually collected from patients are inaccurate and subjective and may not reflect the true status of vitamin B_{12} intake. Second, the information regarding metformin compliance was not collected. Compliance can have an influence on both the response to metformin and the concentrations of vitamin B_{12}. The third limitation is the absence of a control group to abolish the effects of diabetes per se. However, it is considered unethical to recruit diabetes patients who are not receiving intervention. Lastly, we have used only serum vitamin B_{12} level to define vitamin deficiency and have not measured serum holotranscoblamin II, homocysteine and methylmalonic acid which are more sensitive indicators of vitamin B_{12} status.

Large, multicentre studies are recommended on the subject are recommended.

Conclusion

Metformin was found to be associated with vitamin B_{12} deficiency, but this deficiency had no correlation with peripheral neuropathy in T2DM patients.

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Conflict of Interest: Authors have no conflict of interest

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