Research Article

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Vitamin B12 is Found Sufficient in Newly Diagnosed Type 2 Diabetes in a Hospital Based Study

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Abstract

Background: *Vitamin B12 deficiency may be related to peripheral neuropathy in people with type 2 diabetes mellitus (T2DM). Level of B12 in T2DM observed by many investigators showed variable results. Studies on vitamin B12 in T2DM are very limited in Bangladesh.*

Objectives: *To observe serum vitamin B12 level in newly diagnosed T2DM patients.*

Methods: *Observational cross-sectional study encompassing 50 newly diagnosed T2DM and 50 controls as per American Diabetes Association (ADA) criteria. Vitamin B12 and Hemoglobin A1c (HbA1c) were measured for all.*

Results: *Both mean (492.46±28.82 vs. 346.48±19.65 pg/mL, mean±SEM; p=<0.001) and median (435.50 vs. 334.50 pg/mL) values of serum vitamin B12 were found to be higher in T2DM than those of controls. None of the diabetic subjects were found to be B12 deficient whereas 6 were borderline deficient; these frequencies were 7 and 11 respectively among the controls. Vitamin B12 level was statistically similar in patients with or without clinically evident peripheral neuropathy (mean±SEM; 523.48±39.39 vs. 441.84±38.76 pg/mL, p=0.172). B12 level showed positive correlation with fasting plasma glucose (FPG, r=0.285, p=0.061) and HbA1c (r=0.287, p= 0.043) in diabetes group but there was no correlation with body mass index (BMI).*

Conclusion: *Vitamin B12 is found sufficient in newly diagnosed Bangladeshi T2DM patients.*

Keywords: Vitamin B12, Type 2 DM, HbA1c.

Introduction

Diabetes mellitus (DM) is associated with development of micro- and macro-vascular complications and peripheral neuropathy is one of the microvascular complications of DM [1]. Vitamin B12 is a water soluble vitamin [2]. It is essential in the synthesis of neuronal myelin sheath and also in the synthesis of monoamines or neurotransmitters like serotonin and dopamine [3]. Axonal demyelination, degeneration and later death are the hallmark of vitamin B12 deficiency induced neuronal damage that manifest as severe peripheral or autonomic neuropathy, sub acute combined degeneration of the spinal cord, delirium and dementia [4,5]. Various studies have shown increased prevalence of vitamin B12 deficiency in patients with type 2 DM [7,6]. Vitamin B12 was found to be low in obese and even overweight persons with or without metabolic syndrome [8]. Metformin use has been implicated as a cause of vitamin B12 deficiency in patients with diabetes though there are reports that it can happen without treatment with metformin [6,7,9]. Barghouti et al. found that diabetic patients are less likely to have B12 deficiency in comparison to nondiabetic subjects [10].

Peripheral neuropathy caused by DM and vitamin B12 deficiency may produce overlapping clinical pictures [11]. Moreover, nondiabetic neuropathies may be present in patients with diabetes [12]. The relatively high prevalence of B12 deficiency in diabetic patients makes it likely that at least some peripheral neuropathy in diabetic patients may be attributable to B12 deficiency [13]. It has been observed that, independent factors other than glycemic control are critical to the development of diabetic polyneuropathy and strict glucose control is not enough to ameliorate the onset and

progression of T2DM diabetic polyneuropathy [14]. Moreover, several published studies reported that therapeutic supplementation with B12 or vitamin B complex mixtures containing B12 significantly improved symptoms in diabetic neuropathy patients [15-17].

The present study was intended to find out the serum vitamin B12 status in newly diagnosed type 2 DM and also would attempt to correlate the deficiency status, if any, with clinical findings of peripheral neuropathy.

Methods and Materials

Subjects and methods

This observational cross-sectional study was conducted in the department of Endocrinology, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka from March, 2014 to April, 2015 after obtaining ethical approval from Institutional Review Board (IRB), BSMMU. The study recruited 50 newly diagnosed cases of T2DM and 50 controls with normal glucose tolerance (NGT); all were Bangladeshi aged 30-45 years of both sexes (Table 1).

Parameters	T2DM (n=50)	NGT (n=50)	p
Age (years, mean ± SD)	37.52 ± 5.72	32.88 ± 3.00	<0.001
Male	25 (50%)	34 (68%)	0.103
Female	25 (50%)	16 (32%)	
Smoker (%)	9 (18%)	3 (6%)	0.121
Body Mass Index (BMI, kg/m^2)	25.43 ± 4.17	24.36 ± 3.21	0.152
Pulse (beats/min)	84.16 ± 10.26	78.20 ± 7.82	0.002
Systolic BP (mmHg)	122.00 ± 19.06	115.30 ± 9.11	0.028
Diastolic BP (mmHg)	79.20 ± 10.61	75.30 ± 6.95	0.032
Fasting Plasma Glucose (mmol/L)	12.42 ± 4.99 (n=44)	4.86 ± 0.41	<0.001
Plasma Glucose 2 Hours after 75 gm Glucose or Breakfast (mmol/L)	18.91 ± 5.79 (n=36)	5.77 ± 1.08	<0.001
HbA1c (%)	10.69 ± 2.64	5.34 ± 0.35	<0.001

Table 1: Characteristics of participants; T2DM= Type 2 Diabetes Mellitus; NGT= Normal Glucose Tolerance; BP= Blood Pressure by Student's t-test and χ^2-test (as applicable).

Diabetes and NGT status were defined as per American Diabetes Association (ADA) criteria for diagnosis of diabetes in non-pregnant adults [12]. Patients with pre diabetes were discarded. Subjects with current or recent metformin & other antidiabetic drug use; any co-morbid conditions that may interfere vitamin B12 level (e.g. pernicious anaemia, malabsorption); chronic (3 months or more) use of acid suppressants i.e. proton pump inhibitors or H2 receptor blockers; getting supplementation of vitamin B12 or any vitamin B12 containing multivitamin, either by oral or parenteral route; concomitant drugs use that affect vitamin B12 level (e.g. corticosteroids, phenytoin, dihydrofolate reductase inhibitors) were excluded during sampling process. After having informed written consent the participants were interviewed and examined for clinical features of peripheral neuropathy. Data were collected using prescribed data collection sheet. Assay of vitamin B12 and HbA1c of collected samples was done in Department of Biochemistry, BSMMU on the same day of blood sample collection (preferably within 08 hours). Serum vitamin B12 was measured by Chemiluminescent Microparticle Immunoassay (CMIA) technology and HbA1c by High Performance Liquid Chromatography (HPLC) method. Serum creatinine, urinary albumin, haemoglobin and mean corpuscular volume (MCV) were measured in diabetic patients. Serum vitamin B12 ≤200 pg/ml was labeled as deficiency, >200 to ≤300 pg/ml borderline deficiency and >300 pg/ml was labeled as normal [18].

Statistical analysis

Data were analyzed using computer based SPSS program (version 22.0). Results were expressed in frequencies or percentages. Comparison of vitamin B12 level and HbA1c between NGT and DM was done by Student's unpaired t-test. Frequency of reduced vitamin B12 using recommended cut-off values were compared between NGT and DM groups by Chi-square test. Pearson's correlation test was used to observe correlation among different variables (HbA1c, vitamin B12, fasting glucose, age etc). P value ≤0.05 was considered significant.

Results

Table 2 shows the mean and median values of vitamin B12 in T2DM and NGT and figure-1 depicts the frequency of subjects under different status of vitamin B12 made on the basis of recommended cut-off value. There was no statistical difference of vitamin B12 between male and female either in T2DM (473.04 ± 41.59 vs. 511.88 ± 40.39 pg/mL, mean ± SEM; p=0.506) or in control group (334.47 ± 19.07 vs. 372.00 ± 46.69 pg/mL, mean ± SEM; p=0.465).

Group of study subjects	Serum Vitamin B12 (pg/mL)	
	Mean ± SEM	Median
T2DM (n=50)	492.46 ± 28.82	435.50
NGT (n=50)	346.48 ± 19.65	334.50
p	<0.001	
Total (n=100)		

Table 2: Vitamin B12 level in subjects with T2DM & NGT; T2DM= Type 2 Diabetes Mellitus; NGT= Normal Glucose Tolerance; SEM= Standard Error of Mean by Student's t-test.

Table 3 displays vitamin B12, HbA1c, MCV and serum creatinine levels in T2DM subjects with or without peripheral neuropathy. There was found no significant difference between two groups for these variables. Relationships of vitamin B12 with other variables are shown in table 4. Vitamin B12 correlated with FPG (r=0.285, p=0.061) and HbA1c (r=0.287, p=0.043) in T2DM but not in NGT group (r=-0.087, p=0.549 & r=0.003, p=0.983 respectively). BMI did not correlate with vitamin B12 in T2DM (r=-0.232, p=0.105) or in NGT (r=-0.091, p=0.530). As displayed in Table 5, none

among the variables of age, sex, smoking status, BMI, systolic BP, diastolic BP, FPG, 2 hour plasma glucose, HbA1c, Hb, MCV and serum creatinine were found to be independently related to vitamin B12 (p= not significant).

Variables	Neuropathy (n=31)	No neuropathy (n=19)	p
Vitamin B12 (pg/mL)	523.48 ± 39.39	441.84 ± 38.76	0.172
HbA1c (%)	10.76 ± 0.41	10.58 ± 0.72	0.837
MCV (fl	85.58 ± 1.10	84.33 ± 1.76	0.528
S. Creatinine (mg/dL)	0.89 ± 0.04	0.82 ± 0.05	0.309

Table 3: Vitamin B12, HbA1c, and MCV (Mean±SEM) in clinically detectable neuropathic subjects among diabetics (n=50); MCV= Mean Corpuscular Volume; fl= Famtoliter; SEM= Standard Error of Mean Any or more subjective or clinically detectable finding were considered as neuropathy by Student's t-test.

Variables	Type 2 DM (n=50)		NGT (n=50)	
	R	*p*	*r*	*p*
Age and Vit. B12	0.061	0.673	-0.063	0.663
BMI and Vit. B12	-0.232	0.105	-0.091	0.530
FPG and Vit. B12	0.285	0.061	-0.087	0.549
Plasma Glucose 2 Hours after 75 gm Glucose /Breakfast and Vit. B12	0.148	0.388	-0.021	0.886
Hemoglobin and Vit. B12	0.089	0.537		
MCV and Vitamin B12	-0.020	0.890		

Table 4: Correlations; DM= Diabetes Mellitus; NGT= Normal Glucose Tolerance; FPG= Fasting Plasma Glucose; MCV= Mean Corpuscular Volume by Pearson's correlation.

Variables	B	SE	Beta	t	p
(constant)	-2.025	1.573	-1.288	0.211	
Age	0.012	0.014	0.200	0.882	0.387
Sex	0.342	0.196	0.494	1.748	0.094
Smoking status	0.150	0.220	0.150	0.683	0.501
BMI	0.034	0.025	0.339	1.379	0.181
Systolic BP	-0.007	0.006	-0.368	-1.180	0.250
Diastolic BP	0.011	0.009	0.333	1.180	0.250
Fasting plasma glucose	-0.019	0.042	-0.196	-0.449	0.658
Plasma Glucose 2 Hours after 75 gm Glucose/ Breakfast	0.052	0.030	0.860	1.763	0.091
HbA1c	0.001	0.054	0.005	0.011	0.991
Hemoglobin	0.060	0.053	0.313	1.141	0.265
MCV	-0.013	0.012	-0.274	-1.062	0.299
S. Creatinine	0.262	0.384	0.164	0.682	0.502

Table 5: Multiple regressions for serum vitamin B12 status in T2DM; T2DM= Type 2 Diabetes Mellitus; BMI= Body Mass Index; BP= Blood Pressure; MCV= Mean Corpuscular Volume.

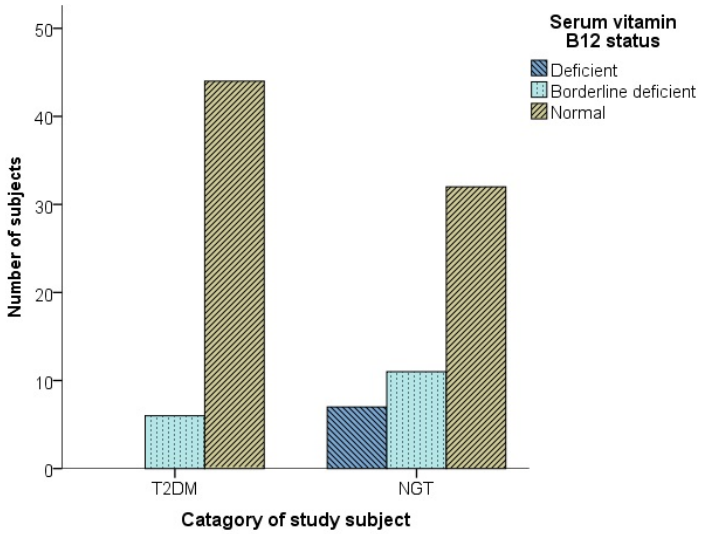

Figure 1: Vitamin B12 status in T2DM & NGT; T2DM=Type 2 Diabetes Mellitus; NGT= Normal Glucose Tolerance.

Discussion

The present study clearly observed that newly detected T2DM subjects had higher serum vitamin B12 than those of NGT and the numbers of subjects with subnormal B12 level (deficient and borderline deficient) were higher in NGT group. None was B12 deficient in T2DM group, but 7 out of 50 NGT fell into deficient group while 6 in T2DM and 11 in NGT fell into borderline deficiency. So, the frequency of subnormal serum B12 was 36% in NGT group in comparison to only 12% in DM group. This finding is similar to a Jordanian study which revealed that diabetic patients were less likely to have B12 deficiency [10]. We did not find any of the T2DM subject to have B12 deficiency and only a few of the control subjects were found to be deficient or borderline deficient. Contrary to our findings, Al-Maskari, et al. found significantly lower levels serum vitamin B12 in newly diagnosed T2DM than NGT controls among Omani people [19]. Another study conducted in Nigeria revealed significantly low vitamin B12 in new diabetics when compared to the controls [6]. Many studies observed that vitamin B12 deficiency is a common but under-recognized disorder with prevalence of 3%-40% in the adult population [20-23]. In this context, it may be relevant to mention that people of Indian subcontinent is reported to have higher prevalence of deficiency [24,25]. One study done in Pune, India discovered that about 75% of the adults had metabolic evidence of B12 deficiency and 47% of adults had low vitamin B12 [25]. Liggy, et al. found 65.6% of Gujrati people to have low B12 levels among those who were advised vitamin B12 test by the clinicians [25].

Present study found no significant difference of B12 level between males and females in either DM or NGT group which is in agreement with study done by Baltaci, et al. who found no significant difference of B12 between obese male and female patients [8]. In contrast, Barghouti, et al. found lower levels of B12 in men compared to women [10]. It was interesting to observe that our study found higher levels of vitamin B12 in diabetic

subjects with clinically suspected peripheral neuropathy than those without neuropathy. This finding is in agreement with the studies conducted by Mørkridet, et al. and Kiani, et al. both of whom found serum vitamin B12 level to be higher in patients with diabetic peripheral neuropathy (DPN) than those without DPN [26,27]. But other studies observed opposite findings. Ambrosch, et al. found that the vitamin B12 concentration demonstrated a trend to decrease in neuropathy group though the frequency of B12 deficiency under cut off value was similar in both groups [28]. Sachedina, et al. observed that B12 deficiency was associated with peripheral neuropathy of greater severity [11].

Our study found no significant correlation of B12 level with BMI either in T2DM or in controls though in both the cases the trend was inversely related. Ebesunum, et al. found no significant correlation of B12 level with BMI in T2DM patients [6]. In contrast to our finding, Baltaci, et al. found that B12 level was significantly lower in patients with obesity and over weight than normal weight individuals and observed significant but inverse correlation between vitamin B12 and BMI [8]. In our study, we did not find any statistical difference of BMI between the two groups of T2DM and control. This may be due to small number of subjects in the present study.

Conclusion

In conclusion, this study found that serum vitamin B12 was sufficient in subjects with newly diagnosed type 2 DM. There was no statistical difference between the sex groups or between subjects with and without clinically suspected neuropathy. However, it needs wide scale studies to understand situation of vitamin B12 in T2DM properly.

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