ORIGINAL ARTICLE

To Determine Mean Reduction in Homocysteine Levels in patients with Type 2 Diabetes Mellitus, on Vitamin B_{12} Therapy

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ABSTRACT

Aim: To determine mean reduction in homocysteine levels with type 2 DM on Vit. B12 therapy.
Study design: Quasi-experimental study.
Study duration: from January 5th 2008 to July 4th 2008
Place: Medicine Unit II Abbasi Shaheed Hospital, Karachi
Patients and method: A total of 72 patients were enrolled during six months study period. Blood samples were taken for fasting blood glucose and fasting homocysteine at the start of study to assess the inclusion criteria. Tab. Vitamin B_{12} 500microgm/day for 6 weeks was given to the patients. Fasting homocysteine levels was again determined after 6 weeks. To ensure minimization of any bias sample collection, transportation, laboratory procedures were kept standardized. Data analysis was done by using statistical package for social sciences
Results: The mean difference in homocystien before and after treatment was 0.39 (95% CI: 0.36-0.42) and is statistically significant p-value <0.001.
Conclusion: It is concluded that vitamin B12 has significant role in reducing homocystein level in patients with type II diabetes.
Keywords: Homocystien levels, type II diabetes mellitus, Vitamin B12,

INTRODUCTION

Diabetes mellitus is a syndrome with disordered metabolism and inappropriate hyperglycemia due to either a deficiency of insulin secretion or a combination of insulin resistance and inadequate insulin secretion to compensate[1,3]. There are two main types of diabetes, Type1 and Type 2. The third group is labeled as "other specific type", by American Diabetics Association (ADA)[2,3].

Homocysteine is an amino acid in the blood, formed by the liver by another aminoacid methionine, found in animals and plants protein. Even though 70% of it is protein bound, it is a potent toxin to endothelial cells. Methyltetrahydrofolate and Cobalamin plays role in its metabolism thus prevent its accumulation[4].

Hyperhomocysteinemia is a risk factor for overall mortality in Type 2 diabetics. For each 5µmol/L increment of serum total homocysteine, the risk of 5 years mortality rose by17% in the non-diabetics and by 60% in the diabetics[1]. Hyperhomocysteinemia may exert an atherosclerotic effect that may lead to cardiovascular disease. Several prospective studies have investigated the relation between total homocysteine and risk of cardiovascular disease. Many[2,3,4,5,6,7,8,9] but not all[10,11,12] found a positive relation between hyperhomocysteinemia and a cardiovascular illness. This atherosclerotic effect is exerted through increased oxidative stress which may induce endothelial dysfunction[13,14,15] Homocysteine can also affect the properties of the extra cellular matrix and smooth muscle cell proliferation[1,19]. Oxidative stress is thought to be increased in type2 Diabetes mellitus[18] and matrix alteration are a prominent feature of diabetes in general, both of which might make diabetic patients more susceptible to the adverse effect of hyperhomocysteinemia.

Serum homocysteine levels are a risk factor for development of nephropathy, without macroalbuminuria at baseline[16]. Homocysteine may be a marker for occult renal damage or that it may cause renal damage per se[16]. An adequate status for folate and vitamin B_{12} is necessary to prevent accumulation of homocysteine in the blood. Recent studies however suggest that large proportion of the population, perhaps 40% is not consuming enough to keep the concentration of homocysteine in plasma low[17].

Subjects taking vitamin B supplements, daily, have low concentration of homocysteine[17] compared with those not taking them, 27% of which had homocysteine level in excess of the reference range (>15µmol/l) and 66% in excess of that recommended by the AHA for at risk groups (>10µmol/l)[20]. Cobalamin supplements were followed by a small but

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significant average decrease in homocysteine concentration. The decrease in homocysteine may thus reflect the use of different derivatives of cobalamin in the intracellular metabolism of homocysteine. Methylcobalamin is an essential cofactor for the enzyme methionine synthase and 5'-deoxy adenosylcobalamin is an essential cofactor for the enzyme methylmelonyl-CoA mutase.[17] Hyper-homocysteinemia if found in type2 diabetes patient may have increase risk of cardiovascular events. If the effect of vitamin B12 in lowering the homocysteine level is proved than it is helpful to the clinicians to advice vitamin B 12 in type 2 diabetes mellitus patients with raised homocysteine, thereby preventing cardiovascular events.

MATERIAL AND METHODS

This Quasi-experimental study was conducted in the Department of Medicine at Abbasi Shaheed Hospital, Karachi. A total of 72 patients were enrolled during six months study period. The type2 diabetic patient for ≥ 10 years of diagnosis, attending OPD and Inpatient Department of Medicine Unit-II, Abbasi Shaheed Hospital, C.D.G.K., Karachi were included. Patients having type1 Diabetes mellitus or having type 2 Diabetes mellitus < 10 years history, patients who are taking vitamin supplements 6 weeks prior to study, patients having terminal illnesses and patients having endocrinal illnesses were excluded from this study. Blood samples were taken for fasting blood glucose and fasting homocysteine at the start of study to assess the inclusion criteria. Tab.Vitamin B_{12} 500microgm/day for 6 weeks was given to the patients. Fasting homocysteine levels was determined after 6 weeks. To minimization of any bias sample collection, transportation, laboratory procedures will be kept standardized. Relevant information including age, gender, fasting blood sugar and fasting homocysteine levels (pre and post vitamin B12intake) was recorded on preapproved proforma.

Data entry and statistical analysis was entered and analyzed using SPSS version 10. Data cleaning was done prior to analysis through frequencies run. Categorical variables (gender) were presented as proportions or percentage; continuous variables (age, FBS, Homocysteine levels) were presented as mean + SD. Mean homocysteine levels was determined at baseline and six weeks after administration of vitamin B_{12}. Pre and post measurements of determine mean serum homocysteine levels was compared by paired t-test and p-value was considered as significant if it is less than or equal to 0.05.

RESULTS

A total of 72 patients were enrolled during six months study period. The mean age of enrolled participants was 60.1±7.3 years (Table 1), 62.5% were male with male to female ratio of 1.7:1 (Table 2). Mean fasting blood sugar was 121.1±6.4 mg/dl (Table 3). Mean homocysteine level before treatment was 14.8±3.1 µmol/L (Table 4) and mean homocysteine level after Vitamin B12 treatment was 14.5±3 µmol/L (Table 5). The mean difference in homocystien before and after treatment was 0.39 (95% CI: 0.36-0.42) and is statistically significant p-value <0.001 (Table 6).

Table 1: Age distribution of enrolled participants (n=72)

Age distribution	Frequency	%age
48-52 years	18	25.0
52.1-60 years	19	26.4
60.1-67 years	21	29.2
67.1-72 years	14	19.4

Mean age: 60.1±7.3 years

Table 2: Sex distribution of enrolled participants (n=72)

Sex	Frequency	%age
Male	45	62.5
Female	27	37.5

Male to female ratio: 1.7:1

Table 3: Frequency of fasting blood sugar in enrolled participants (n=72)

Fasting blood sugar	Frequency	%age
116.1-123 mg/dl	22	30.6
123.1-127 mg/dl	22	30.6
127.1-129 mg/dl	10	13.9

Mean fasting blood sugar: 121.1±6.4 mg/dl

Table 4: Frequency of homocystien level before vitamin B12 treatment in enrolled participants(n=72)

Homocystien level before treatment	Frequency	%age
<13 µmol/L	20	27.8
13-15 µmol/L	32	44.4
>15 µmol/L	20	27.8

Mean homocystien before treatment: 14.8±3.1 µmol/L

Table 5: Frequency of homocystien level after vitamin B12 treatment in enrolled participants(n=72)

Homocystien level after treatment	Frequency	%age
<13 µmol/L	26	36.1
13-15 µmol/L	29	40.3
>15 µmol/L	17	23.6

Mean homocystien after treatment: 14.5±3 µmol/L

Table 6: Mean difference in homocystien level before and after vitamin b12 treatment

	Mean	Std. deviation	95% CI of the difference	
			Upper	Lower
HCY* µmol/l (before)-HCY (after)	0.39	0.14	0.36	0.42

P value: <0.001, **HCY=Homocystien level*

DISCUSSION

Hyperhomocysteinemia is a recently recognized modifiable risk factor for cardiovascular disease that is independent of major risk factors such as diabetes, hypertension, hypercholesterolemia, and smoking[21-22]. The prevalence estimates of hyperhomocysteinemia (>14µmol/L) vary between 5% and 30% in the general population[23-24]. A meta-analysis[25] showed that treatment with 0.5 to 5.0mg folic acid daily can lower serum total homocysteine (tHcy) by 15% to 40% within ≈6 weeks. In addition, it has been estimated that lowering tHcy by 5 µmol/L (≈1 SD) may reduce the risk of cardiovascular death by ≈10%[26]. Taken together, hyperhomocysteinemia may be an important modifiable risk factor, although this must be confirmed in randomized studies of homocysteine-lowering treatment.

In a cross-sectional analysis, hyperhomocysteinemia appeared to be a stronger risk factor for cardiovascular disease in type 2 diabetic subjects than in nondiabetic subjects[24]. Such an interaction between hyperhomocysteinemia and type 2 diabetes with regard to cardiovascular risk may be clinically important, as it implies that homocysteine-lowering treatment may be especially effective in type 2 diabetes.

Vitamin B_{12} deficiency is traditionally diagnosed by laboratory findings of low serum vitamin B_{12} levels, typically in the setting of megaloblastic anemia. However, subclinical B_{12} deficiency often presents with normal serum B_{12} levels and hematologic parameters[27]. Elevated methylmalonic acid and homocysteine levels improve the diagnosis of tissue B_{12} deficiency[28-29] and may identify patients with deficiency at an early, reversible stage. Using these more specific diagnostic markers, we conducted a cross-sectional study to determine the extent of B_{12} deficiency in the diabetic population.

In this study we found that mean homocystien level before treatment was 14.8±3.1µmol/L and mean homocystien level after Vitamin B12 treatment was 14.5±3 µmol/L. The mean difference in homocystien before and after treatment was 0.39 (95% CI: 0.36-0.42) and is statistically significant p-value <0.001.

Those who were taking vitamin B supplements, have low concentration of homocysteine[17] compared with those not taking them, 27% of which had homocysteine level in excess of the reference range(>15µmol/l) and 66% in excess of that recommended by the AHA for at risk groups (>10µmol/l).

CONCLUSION

It was concluded from this study that Vitamin B12 supplementation significantly lowers the homocystein levels in type II diabetic patients. Hyperhomocysteinemia is modifiable risk factor of non-communicable diseases as whole and through this simple and cost effective measure.

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