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Comparative study of role of Macro minerals & Electrolytes with Atherogenic index of plasma in Diabetic men and women

Neha Sharma[1]*, Umesh Kumar pareek[1], Avdhesh kumar Sharma[1], Ketan Mangukiya[1], Mali KL[1], Kaushik GG[2]

1. Geetanjali Medical College & Hospital, Udaipur, Rajasthan, India
2. J.L.N. medical college & Hospital, Ajmer, Rajasthan, India

***Corresponding Author:** Neha Sharma, Assistant Professor, Department Of Biochemistry, Geetanjali Medical College & Hospital, Udaipur, Rajasthan, India; Email : neha16.sharma@gmail.com

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ABSTRACT

The prevalence of diabetes in developing country is increasing rapidly. However, scarce data are available on serum electrolyte levels and macro metal calcium and magnesium in adults with diabetes, especially in those with cardiovascular complications. This study measured serum electrolyte levels, serum calcium, serum magnesium, glycosylated hemoglobin and examined their relationship with macro vascular complications in adults with diabetes in Indian population. Comparison of both men and women (men= 290, women=110) in the age group of 23 to 70 years were enrolled, Fasting plasma glucose, glycol Hb, AIP, calcium, magnesium and electrolytes were measured using the fasting blood samples. Serum sodium and magnesium levels in the diabetes group were significantly decreased, while the serum calcium level was significantly increased. Serum calcium level was positively correlated with HbA1c. The study recommends that along with glucose and Glycosylated hemoglobin, calcium and magnesium, AIP, electrolytes assays too should be carried out as additional parameters in the evolution of diabetes mellitus and macro vascular complications especially for Diabetic male patients between age group 46-70 years with or without any previous history of CVD.

Key words: Electrolytes, Macro vascular complications, Diabetes mellitus, Fasting Plasma Glucose, Glycosylated Hemoglobin, Calcium, Magnesium, Atherogenic index of plasma.

Abbreviations' Used: CVD ; cardio vascular diseases., AIP; atherogenic index of plasma, TC ; total cholesterol, TG; triacylglycerol, HDL ; high density lipoprotein, LDL ; low density lipoprotein, BMI; body mass index, FBS; fasting blood glucose, PPBS, post parandial blood glucose.

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1. INTRODUCTION

Diabetes can cause many complications. Acute complications (hypoglycemia, ketoacidosis, or nonketotic hyperosmolar coma) may occur if the disease is not adequately controlled. Serious long-term complications include cardiovascular disease (doubled risk), chronic renal failure, retinal damage (which can lead to blindness), nerve damage (of several kinds), and microvascular damage, which may cause impotence and poor healing. Poor healing of wounds, particularly of the feet, can lead to gangrene, which may require amputation. Diabetes mellitus is characterized by recurrent or persistent hyperglycemia, and is diagnosed by demonstrating any one of the following: · fasting plasma glucose level at or above 126 mg/dL (7.0 mmol/l). Plasma glucose at or above 200 mg/dL (11.1 mmol/l) two hours, after a 75 g oral glucose load as in a glucose tolerance test, random plasma glucose at or above 200 mg/dL (11.1 mmol/l), (WHO report, 2006).

Most physicians prefer to measure a fasting glucose level because of the ease of measurement and the considerable time commitment of formal glucose tolerance testing, which takes two hours to complete. According to the current definition, two fasting glucose measurements above 126 mg/dL (7.0 mmol/l) is considered diagnostic for diabetes mellitus (Saunders, 1995).

Diabetes Mellitus comprises a heterogeneous group of diseases, which causes the patients, if untreated to have abnormally high plasma glucose levels. Magnesium is an important element for health and disease. It is the second most abundant intracellular cation; has been identified as a cofactor in over 300 enzymatic reactions involving energy metabolism, protein and nucleic acid synthesis. Approximately 50% is in soft tissues, the other half in bone and less than 1 % is present in blood. The clinical laboratory evaluation of magnesium status is limited to the level of magnesium in serum, 24 hour urinary excretion and percentage retention following parenteral administration (De Valk, 1999). Diabetic nephropathy and other complications are most likely to be worse as a result of concurrent Magnesium deficiency (Fox, 1999).

In prospective studies, low calcium intake is consistently found to be inversely associated with incident type 2DM or the metabolic syndrome. In the Nurses' Health Study, total (food + supplements) calcium intake was inversely associated with incident type 2DM after complete multivariate adjustment, including vitamin D intake. Ca^{2+} is mainly reabsorbed in the proximal tubule. Its reabsorption is coupled to Na^{+} absorption, and it appears to compete with Mg^{2+} for transport in the loop of Henle. In the distal convoluted tubule, Ca^{2+} absorption is regulated independently of Na^{+}, where numerous factors, such as calcitonin, parathyroid hormone, and vitamin D, can have marked effects on Ca^{2+} reabsorption and secretion (Hall and Guyton, 2006; Vallon et al. 1999). Energy metabolism is associated with bone remodeling, and glucose levels also regulate parathyroid hormone and vitamin D levels (Agus, 1999).

The electrolytes in serum include sodium (Na^{+}), potassium (K^{+}), calcium (Ca^{2+}) and magnesium (Mg^{2+}) (Frost et al. 2010). These electrolytes play an important role in intermediary metabolism and cellular function, including enzyme activities and electrical gradients. Serum concentrations of electrolytes have been shown to change with plasma glucose levels (De Fronzo, 1976). Disturbances in the levels of some electrolytes are associated with diabetes mellitus (DM), (Katz, 1973; Al-Rubeaanet al. 2011; Levy et al. 1986; Simmons et al. 2010). The Glycohemaglobin test is one blood sample every 3 to 4 months, and the test does not change with any recent changes in diet, exercise, or medicines. Two main functions of Glycosylated hemoglobin viz HbA1 or HbA1C are commonly used in diabetes monitoring.

The purpose of the present study was to retrospectively evaluate the association between serum electrolytes, calcium magnesium and glycosylated hemoglobin with AIP in type 2 diabetes male and female patients with or without complain of cardiac diseases.

2. MATERIAL AND METHODS

This study was carried out in Geetanjali hospital and medical college. This was a retrospective study which was conducted on 400 diabetic subjects between March 2014 to September 2014. All the subjects for study were in age group between 23-70 years. All data for the study was obtained from the clinical biochemistry laboratory of Geetanjali hospital.

The subjects were divided in to men (290) and women (110). Further according to age group and clinical history they were divided into four groups. Men with history of CVD and without history of CVD in age group 23-45 (n=73; 23, 50) and 46-70 (n= 217; 97, 120) years, respectively. Similar women were also divided in to the group women with history of CVD and without history of CVD, in age group 23-45 (n=20; 5, 15) and 46-70 (n=90; 25, 65) years respectively.

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Inclusion criteria of the study were diabetic subjects with or without cardiac complain, between age group 23-70 or not having any complained of severe illness. Exclusion criteria of the retrospective study were the subjects with any other clinical history instead of Diabetes and heart diseases.

Height and weight of subjects were measured by standard protocol. Body mass index (BMI) was calculated. For the estimation of Serum calcium, Magnesium, lipid profile and electrolytes, 5 ml serum were taken by standard protocol. Serum glucose, serum triglyceride and serum total cholesterol were analyzed spectrophotometrically by GOD-PAP, GPOPAP and CHOD-PAP methods respectively by employing reagent kits on a fully automated analyzer of the Cobas c-311 series. Atherogenic index of plasma was calculated as log (TG/HDL-C) with TG and HDL-C expressed in molar concentrations. It has been suggested that AIP value of −0.3 to 0.1 is associated with low CAD risk, 0.1 to 0.24 medium and above 0.24 high risks. Serum calcium level were analysed by NM-BPTA (N-methy-bis Aminophenoxy ethane tetra acetic acid) method. Serum electrolytes were measured by roche9180 electrolyte analyzer. Serum magnesium measured by colorimetric end point method based on reaction of magnesium with xylidyl blue in alkaline solution containing EGTA to mask the calcium in the sample.

Determination of HbA1c

For the estimation of glycosylated hemoglobin whole blood sample was collected and measured by cobas c- 311 immunoturbidometric Gen -2 whole blood acc to IFCC method.

Principle

Both the concentration of HbA1c and the concentration of total hemoglobin are measured. The reported HbA1c result is calculated as a % of total hemoglobin concentration. The HbA1c and total hemoglobin values generated in this assay are intended for use in the calculation of the HbA1c/total hemoglobin ratio (% HbA1c) and must not be used individually for diagnostic purposes.

Sample pre-treatment

The first step of the procedure involves the pre-treatment of the whole blood sample. This lyses red blood cells and causes hydrolysis of the hemoglobin by the action of a protease enzyme in the hemoglobin denaturant reagent.

Statistically analysis

Results are presented as mean ± SD. A p value < 0.05 was considered significant. Statistical analysis was performed using GraphPad Prism version 5.00 for Windows (GraphPad software, San Diego California USA, www.graphpad.com).

3. RESULTS

Hypomagnesaemia and decreased sodium has been linked to poor glycemic control, coronary artery diseases, hypertension, diabetic retinopathy, and nephropathy, neuropathy, and foot ulcerations**.** In this study total subjects were 400. They were further divided in different group according to their age sex and any history of cardiac disease. In this study we were found that there were no significant changes in Body mass index, while in all group instead of men between age group 46-70 year fasting blood sugar were statically significant, in the similar manner postprandial blood glucose were significant in age group between 46-70 years while it was no significant for men /women between age group 23-45 years with or without history of CVD.

Atherogenic index of plasma is significant only in men with and without history of CVD between 46-70 years, while it was no significant for rest subjects between 23-45 years. Higher risk of CVD along with the changes in macro metals and electrolytes were found in Men between age group 46-70 years in comparisons to women.

Table 1 shows the distribution between different Age and sex with or without history of CVD. Table 2 represents FBS, PPBS, AIP and HbA1C of all groups, table 3 represent Difference in TC, TG, HDL and electrolytes. Table 4 and 5 shows the p- value of subjects. Diabetic macro vascular complications, CVD in particular, are currently the leading cause of death for diabetic patients.

4. DISCUSSION

Derangement of water and electrolyte balances may occur in subjects with DM, resulting from insulin deficiency, hyperglycemia and hyperketonemia (Hatwal et al. 1989). Significant reductions in serum Na^+ and Mg^{2+} levels and an elevation in serum Ca^{2+} in subjects with DM were observed. These results were consistent with those reported by previous studies (Vallon et al. 1999; Agus, 1999). Many studies have shown that both mean plasma and intracellular free magnesium levels are lower in patients with diabetes than in the general population. This magnesium deficiency, which may take the form of a chronic latent magnesium deficit rather than clinical hypomagnesaemia, may have

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clinical importance because the magnesium ion is a crucial cofactor for many enzymatic reactions Involved in metabolic processes (Rodriguez-Moran and Guerrero-Romero, 2001).

In our study we were found a significant change in magnesium between age group 46-70 years in both men and women. Our study was correlated with other study. Magnesium deficiency is associated with insulin resistance and increased platelet reactivity, but studies of oral magnesium supplementation and changes in glycemic status or lipid levels in diabetes have not been conclusive. Oral magnesium to 20 patients with type 2 diabetes, intracellular free magnesium concentration in erythrocytes normalized and the increase in platelet reactivity in response to thromboxanes decreased significantly (Vallon et al. 1999).

Diabetic macro vascular complications in this study included any patient with a history of CVD events. In our study we were find a negative correlation between Serum sodium level and glucose. While decreased the level magnesium in diabetic patients with and without history of CVD. In this study increased the level of calcium in the Diabetic patients with history of CVD in comparisons to without history of CVD. Change in this level was very in men more than women. Status of serum potassium in patients with history of CVD was increased in comparisons to those who were having no any previous history of CVD in Diabetic Subjects (Hatwal et al. 1989).

The role of calcium in the development of type 2 diabetes is suggested indirectly by cross-sectional studies in which high calcium intake has been found to be inversely associated with body weight and fatness (Hatwal et al. 1989: Rodriguez-Moran and Guerrero-Romero, 2001; Dobiásová and Frohlich, 2001). The results from small clinical trials have been inconsistent (Kitabchi et al. 2006; McNair et al. 1982; Haglin et al. 2011; Kao et al. 1999). Furthermore, levels of serum Na^{+} and Mg^{2+} were negatively correlated with plasma glucose levels, measured as FPG, 2hPG and HbA1c, while Ca^{2+} level was positively correlated with HbA1c in subjects with DM. Intracellular Ca^{2+} plays a critical role in muscle contractions, insulin secretion, and glucose uptake after the binding of insulin to muscle cell membranes (Bjornholm and Zierath, 2005).

In our study a significant change were occurred in men between 46-70 years in comparisons to women in calcium and sodium. No significant changes were present in PPBS in men /women between 23-45 years with or without history of CVD. The most common clinical manifestations of type 2 DM is insulin resistance, which mostly occurs in skeletal muscle, and the impairment of insulin secretion (Paolisso et al. 1990). All these factors may account for the elevation of serum Ca^{2+} level in subjects with DM, and especially in patients whose eGFR levels are increased. Both decreased intracellular and extracellular Mg^{2+} concentrations may accelerate atherogenesis via the elevation of inflammatory cytokines and lipid oxidation, an increase in endothelial cell growth and the inhibition of cellular DNA repair (Hashimoto et al. 2010; Maier et al. 2004; Adhikari et al. 2006).

Diabetic macro vascular complications, CVD in particular, are currently the leading cause of death for diabetic patients. It has been reported that even low Mg^{2+} levels that are still within the normal reference range are associated with all-cause mortality in patients with T2DM (WHO expert Committee on Diabetes Mellitus, 1979).

5. CONCLUSION

Concluded that changes in macro metals and electrolytes were more significant in male between age group 46-70 years in comparisons to Women/ men between age group 23-45 years. Higher risk of vascular complications along with the changes in macro metals and electrolytes were found in Men between age group 46-70 years in comparisons to women.

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DECLARATION OF INTEREST

No declaration of interest.

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Table 1

Distribution of subjects according to age & sex with clinical history

Total (Men=290, women,110)	Age /Sex 23-45 years	Age / Sex 46-70 years	Percentage(23-45 years)	Percentage(46-70 years)
Men with CVD History	23	97	7.93%	33.44%
Men without CVD history	50	120	17.24%	48%
Women with CVD history	5	25	4.5%	22.72%
Women without CVD history	15	65	13.63%	59.09%

Table 2

Comparisons of anthropometric parameter and biochemical parameters

Subjects	BMI	FBS	PPBS	HbA1C	AIP
Age 23-45 years	Mean ±Sd	Mean ± Sd	Mean ±Sd	Mean ± Sd	Mean ± Sd
Men With History of CVD	24.±3.37	210 ± 20.42	250±24.7	10.47 ±1.24	0.123 ± 0.321
Men Without History of CVD	25 ±3.87	198 ± 18.7	248 ±20.22	9.87 ±0.98	-0.110 ± 0.279
p-value	**0.2899 Ns**	**0.01575S**	**0.7157 Ns**	**0.0288 S**	**0.8606 Ns**
Women With History of CVD	25 ±4.1	230±19.8	267 ±25.2	10.56 ±1.11	0.121 ± 0.311
Women Without history of CVD	24.2±3.92	212 ± 18.5	254 ±22.7	8.7± 0.87	-0.114 ± 0.269
p-value	**0.7003 Ns**	**0.0801 S**	**0.2938 Ns**	**0.011 S**	**0.9618 Ns**
Age 46-70 years					
Men With History of CVD	24.22 ±3.58	213 ±20.3	247 ± 21.2	10.78 ±2.1	0.122 ± 0.320
Men Without History of CVD	25.2 ±4.0	216 ±22.5	256 ±21.5	8.56 ±1.1	-0.107 ± 0.259
p-value	**0.0615 NS**	**0.3090 NS**	**0.0023 S**	**0.0001 S**	**0.0081S**
Women With History of CVD	26±3.1	235 ±23.2	275 ±25.22	11.01 ± 1.98	0.125 ± 0.319
Without History of CVD	25.7 ±4.5	205 ±20.8	234 ±20.34	9.56 ±0.98	-0.105 ± 0.275
p-value	**0.7416Ns**	**0.0001S**	**0.0001 S**	**0.0001 S**	**0.7300 Ns**

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Table 3

Comparisons of anthropometric parameter and biochemical parameters

Subjects	TC Mg/dl	TG Mg/dl	HDL Mg/dl	Na+ Mmol/l	K+ Mmol/l	Ca++ Mg/dl	Mg++ mg/dl
Age 23-45 years	Mean ± Sd	Mean ± Sd	Mean ± Sd	Mean ± Sd	Mean ±Sd	Mean ± Sd	Mean ± Sd
Men with CVD History	262 ±11.30	175 ±7.82	47 ±14.42	132 ±2.2	5.1±0.36	10.9 ± 0.39	0.9 ± 0.23
Without CVD history	213 ±10.9	123 ±10.2	50 ±15.12	139± 2.4	4.2 ±0.62	9 ±0.41	1.2 ±0.98
p- value	**0.0001 S**	**0.0001 S**	**0.4271 NS**	**0.0001 S**	**0.0001**	**0.0001 S**	**0.1529 NS**
Women with CVD history	250 ±10.9	155 ±10.9	43 ±14.48	129 ±1.9	5.2 ± 0.45	8.5± 0.76	1.3±0.42
Without CVD history	211 ± 9.7	125 ±10.4	46 ± 14.98	131 ±2.1	3.7±0.42	8.2 ±0.97	1.7±0.45
p- value	**0.0001 S**	**0.0001 S**	**0.7006 Ns**	**0.0760 NS**	**0.0001 S**	**0.5389 Ns**	**0.0978Ns**
Age 46-70 years							
Men with CVD history	245 ± 10.23	169 ± 9.91	41 ±15.23	138 ±2.5	5.8± 0.98	11.4 ±0.79	1.1±0.32
Men without CVD history	210 ± 11.21	146 ± 10.21	64 ±16.60	142 ±1.98	3.9±0.45	10.4±0.42	2.1±1.1
p- value	**0.0001 S**	**0.0001 S**	**0.0001 S**	**0.0001 S**	**0.0001 S**	**0.0001 S**	**0.0001 S**
Women With CVD history	243 ± 10.98	164 ± 9.76	39 ±12.56	125±1.56	5.5 ±1.1	9.1±0.56	0.87±0.44
Women Without CVD history	229 ±11.23	156 ±10.2	52 ±15.45	135 ±2.2	3.8±0.91	8.45 ±0.59	1.9 ±0.78
p- value	**0.0001 S**	**0.0011 S**	**0.0003 S**	**0.0001 S**	**0.0001 S**	**0.0001 S**	**0.0001 S**

Table 4

Comparative P- value of subjects

Subjects	TC	TG	Na+	Ca++	Mg++
P-value Men/women with CVD(23-45 years)	0.0399 **S**	0.0001 **S**	0.0091 **S**	0.0001 **S**	0.0056 **S**
Men/women without CVD(23-45 years)	0.5257 **NS**	0.5097 **NS**	0.0001 **S**	0.0001 **S**	0.0609 **Ns**
Men/women with CVD 46-70 years)	0.3947 **NS**	0.0259 **S**	0.0001 **S**	0.0001 **S**	0.0038 **S**
Men /women Without CVD (46-70 years)	0.0001 **S**	0.0001 **S**	0.0001 **S**	0.0001 **S**	0.1956 **Ns**

S= significant, NS = non significant

Table 5

Comparative P- values of Subjects

Subjects	HDL	K^+
P-value Men/women withCVD(23-45 years)	0.5791 **NS**	0.5937 **NS**
Men/women without CVD(23-45 years)	0.3713 **NS**	0.0048 **S**
Men/women with CVD 46-70 years)	0.5459 **NS**	0.1858 **NS**
Men /women Without CVD (46-70 years)	0.0001 **S**	0.3184 **NS**

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