ANTIOXIDANT VITAMIN STATUS IN TYPE 1 DIABETES MELLITUS PATIENTS

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ABSTRACT

Objective: *The study was designed to determine and compare the levels of both water soluble and fat soluble naturally occurring antioxidant vitamins, in order to know the antioxidant vitamin status of the type 1 diabetic and non-diabetic healthy individuals.*

Material and Methods: *This case control study was conducted at Department of Chemistry, University of Peshawar from November 2004 to August 2005. Blood glucose, triglycerides, total cholesterol, HDL-c, LDL-c, VLDL-c and antioxidant vitamins (ß-Carotene, A, E, and C) levels were determined in 30 Type 1 diabetic and 30 healthy subjects.*

Results: *Values of blood glucose and lipid parameters were observed to be significantly increased (P 0.001) with exception of HDL-c, which was significantly decreased (P 0.001) in Type 1 diabetic patients. The concentration of antioxidant vitamins (ß-carotene, E and C) were found to be significantly low in Type 1 diabetics when compared with normal healthy controls.*

Conclusion: *Observation of low levels of antioxidant vitamins suggests possibly significant defects in antioxidant protection in Type 1 diabetes mellitus patients.*

Key Words: *Type 1 Diabetes Mellitus, Antioxidant Vitamins.*

INTRODUCTION

Diabetes mellitus is defined as a disease of abnormalities of fasting or postprandial glucose and it is frequently associated with disorders of eyes, kidneys, nerves, and circulatory system. [1] The prevalence of diabetes mellitus is increasing worldwide and by the year 2010 it would have affected up to 218 million people. [2] One of the surveys conducted by WHO estimated that by the year 2025, Pakistan will have about 14.5 million people with diabetes. [3]

Type 1, or insulin dependent diabetes mellitus, is considered widely as an organ specific autoimmune disease that leads to destruction of the pancreatic ß-cells. [4-6] The symptoms appear abruptly resulting in absolute lack of insulin. [7,8] Type 1 diabetes mellitus occurs usually in childhood. The tendency for hyperglycemia is life long and also a great risk of developing serious complications. [9] There is also accompanied oxidative stress in diabetes, which is seen as lipid peroxidation, due to increased oxygen free radical production. [10] Increased production of oxygen free radicals have been implicated in the pathogenesis of diabetic complications [11-13] including both microvascular and macrovascular dysfunction. [12-13] Free radical reactions occur in many biological processes. In all aerobic organisms molecular oxygen readily accepts an electron, leading to the formation of oxygen free radicals. [14] The detrimental effects of oxygen free radicals on biological tissues that mediate to their injury [15], are taken care off by the body's defense system using enzymatic mechanisms-superoxide dismutases, catalase, glutathione peroxidase as well as non-enzymatic several antioxidant vitamins. [14] Currently a lot of interest has been shown by the scientific and public communities to elucidate the possible role of antioxidant vitamins in particular vitamin E, C, and beta-carotene, in preventing lipoprotein oxidation and to antagonize the atherosclerosis process. [16]

Considering all this, in the present study we determined the levels of both water soluble and

BLOOD GLUCOSE AND LIPID PROFILE OF CONTROL SUBJECTS AND TYPE-1 DIABETIC INDIVIDUALS

	Control (n=30)	Type-I Diabetes Mellitus (n=30)
Glucose (mg/dl)	91.87 ± 6.32	201.10 ± 46.64 *
Triglycerides (mg/dl)	131.70 ± 13.29	174.70 ± 30.09 *
Cholesterol (mg/dl)	179.53 ± 23.68	225.47 ± 30.66 *
HDL-c (mg/dl)	51.87 ± 8.74	34.83 ± 4.63 *
LDL-c (mg/dl)	101.37 ± 26.80	155.62 ± 27.18 *
VLDL-c (mg/dl)	26.33 ± 2.73	35.00 ± 6.10*

* P 0.001 as compared to Control subjects.

Table 1

ANTIOXIDANT VITAMIN LEVELS OF CONTROL SUBJECTS AND TYPE-1 DIABETIC INDIVIDUALS

	Control (n=30)	Type-I Diabetes Mellitus (n=30)
ß-Carotene (mg/l)	0.99 ± 0.46	0.69 ± 0.28*
Vitamin A [Retinol] (mg/l)	0.47 ± 0.10	0.46 ± 0.11 ¶
Vitamin E [-Tocopherol] (mg/l)	11.73 ± 2.40	8.43 ± 1.43 *
Vitamin C [Ascorbic Acid] (mg/dl)	10.10 ± 1.90	7.27 ± 1.98 *

* P 0.001 as compared to Control subjects.

¶ P value non-significant as compared to Control subjects.

Table 2

fat soluble naturally occurring antioxidant vitamins, in order to know the antioxidant vitamin status of the type 1 diabetic and non-diabetic healthy individuals.

MATERIAL AND METHODS

This study was conducted at Department of Chemistry, University of Peshawar from November 2004 to August 2005. The case control study included 30 patients suffering from type 1 diabetes mellitus and equal number of control healthy individuals. Control subjects having no history of diabetes mellitus, coronary heart disease, hypertension or any other disease participated in this study. The type 1 diabetic patients and control subjects who were on lipid lowering medication and those on multivitamins, especially on antioxidant vitamins were excluded.

Venous blood samples were obtained from all patients and control after overnight fasting. Glucose determination was done immediately after serum was separated from the blood. The serum left was stored at ? 20C for other parameters to be analyzed later. Blood glucose, triglycerides, cholesterol and HDL-c were analyzed by enzymatic colorimetric method, using kits supplied by BioSystems, Spain. LDL-c and VLDL-c were calculated by Friedewald's formula [17] and Wilson's formula [18] respectively. Serum retinol and ß-carotene were determined by using the analytical method proposed by Bradley and Hornbeck [19], whereas serum a-tocopherol assay was done by method recommended by Baker and Frank [19]. Ascorbic acid in the sample reduces 2, 6-dichlorophenol-indophenol, a dye, from blue color to colorless form. When excess dye is added to a solution containing ascorbic acid the decrease in color, determined in a spectrophotometer, is a measure of the amount of ascorbic acid present [20].

Data is expressed as mean ± SD. Statistical significance was evaluated by students t-test. Differences were considered significant at $P<0.05$.

RESULTS

The mean age was 34.47 ± 5.35 years in type 1 diabetes mellitus group (15 female, 15 male) and 42.40 ± 9.89 years in the control group (11 female, 19 male). A statistically significant ($P< 0.001$) difference was observed in the mean age of the study groups, whereas the mean values of BMI in case of type 1 diabetes mellitus group (21.73 ± 3.72 Kg/m^2) and control group (22.97 ± 1.67 Kg/m^2) were indifferent statistically. Comparison of serum glucose and lipid profile between type 1 diabetes mellitus patients and healthy control subjects is shown in Table 1. Fasting serum glucose and the lipid profile were found significantly high ($P< 0.001$), on the other hand the levels of HDL-C was found to be significantly lower ($P< 0.001$) in type 1 diabetes mellitus patients as compared to control group. The levels of beta-carotene, ascorbic acid and alpha-tocopherol were found to be significantly reduced in type 1 diabetes mellitus patients as compared to control subjects. No significant change was observed in the level of serum retinol in type 1 diabetes mellitus patients upon comparison with control subjects (Table 2).

DISCUSSION

Mortality in individuals having diabetes

mellitus is higher than in those people without diabetes, however it varies depending on location and the specific group of people studied [21]. Recent epidemiological studies have pointed out that the incidence of type 1 diabetes mellitus is comparable in children and adults, as this was considered mostly as a childhood disease. [9, 22] Diabetic individuals have a greater risk of developing atherosclerotic disease. This is in part due to the association of diabetic patient with other atherosclerotic risk factors. [23,24] The atherosclerotic vascular disease occurs earlier in diabetics and with greater severity than in non-diabetic subjects. [25] One of the factors that lead to the development of atherosclerosis, are the free oxygen radicals, which participate in this process via lipid peroxides. [26] Individuals with type 1 diabetes mellitus have their lipid profile highly dependent on glycemic control. Those having a poor control show high total triglyceride and cholesterol levels with varying concentrations of high-density lipoprotein cholesterol in comparison with non-diabetic control subjects. On the other hand those diabetic with well-controlled type 1 diabetes mellitus show almost similar or sometimes more favorable, lipid and lipoprotein concentrations than the non-diabetic individuals [27]. This present study reveals increased concentration of triglycerides, total cholesterol, low-density lipoprotein cholesterol and very-low-density lipoprotein cholesterol, whereas the high-density lipoprotein cholesterol levels were significantly lowered in Type1 Diabetes Mellitus patients as compared to the healthy non-diabetic control subjects. Khepsheulidze PN[28] observed elevated concentrations of total cholesterol and low-density lipoprotein cholesterol and lowered levels of high-density lipoprotein cholesterol, while differences in triglycerides and very-low-density lipoprotein cholesterol were insignificant among Type1 Diabetic and non-diabetic healthy children. Torres-Tamayo M et al [29] noticed increased triglycerides and total cholesterol levels in Type1 Diabetic patients. A group of research workers also reported significantly higher serum total cholesterol and apolipoprotein B levels in Type1 Diabetics. [30] Gribauskas PS et al[31] found significantly lower serum retinol concentrations in patients with Type1 Diabetes Mellitus as compared to control subjects. A number of studies [32-36] reported significantly decreased serum retinol levels in younger Type1 Diabetics. Where as, Hozumi M et al [37] showed no significant difference in plasma retinol levels in children with Type1 Diabetes Mellitus in comparison to the healthy controls. The results of our study also show non-significant change in serum retinol levels in Type1 Diabetes Mellitus as compared to control subjects. Beta-carotene levels in the present study were significantly decreased in type 1 diabetics as compared to control group. Granado F et al [33] and Hozumi M et al [37] reported significantly higher serum beta carotene levels in Type1 Diabetes Mellitus patients than normal healthy non-diabetic subjects. Merzouk S et al[34] observed that vitamin C levels were not significantly different between control and insulin dependent diabetic subjects. A number of independent research groups [38-40] reported significantly decreased ascorbic acid concentrations in Type 1 patients when compared to healthy control subjects. Will et al [41] reported significantly lower ascorbic acid levels in persons with newly diagnosed diabetes mellitus than those without diabetes. We also found significantly decreased ascorbic acid levels in Type1 Diabetic patients as compared to control subjects which is in agreement with the afore mentioned studies. Serum alpha-tocopherol levels in our study were significantly decreased in Type1 Diabetes Mellitus. Gribauskas PS et al[31]; Merzouk S et al[34]; Volchegorskii IA [42] found significantly lower levels of serum alpha- tocopherol than normal control healthy subjects. Martinoli L et al [32] and Granado F et al [33] reported no change in the levels of serum alpha-tocopherol in control and insulin dependent diabetic patients. The results of the present study are in agreement with the results of the aforementioned research workers.

CONCLUSION

The basic goal of determination of antioxidant vitamins in type 1 diabetes mellitus patients is to find out the antioxidant status because of the implication of free radical oxidative damage in the pathogenesis of atherosclerotic vascular disease. These findings show significant defects of antioxidant protection in type 1 diabetic individuals, and this may make them more vulnerable to oxidative damage and at a latter stage development of diabetic complications.

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