Original Article

Biochemistry Section

Evaluation of the **Thyriod Status, Oxidant Stress and Antioxidant Status** in patients with **Type - 2 Diabetes Mellitus**

KEDARI G S R

ABSTRACT

Diabetes mellitus is the world's most common endocrine disorder and the aim of the present study was to evaluate the role of the thyroid hormone status in type-2 diabetes cases by measuring serum free 3, 5, 3'-tri-iodothyronine (FT3), free tetra-iodothyronine (FT4) and thyroid stimulating hormone (TSH) levels, the role of oxidative stress by assessing plasma malondialdehyde (MDA) levels, as well as the status of antioxidants like ascorbic acid and reduced glutathione (GSH) in blood. For this, 30 cases of diabetes mellitus were included .The findings were compared with 30 age matched healthy controls, irrespective of sex. A significant increase in the levels of MDA was observed in the cases as compared to the controls. A significant decrease in the levels of serum FT3, vitamin-C, and GSH was also noticed in the cases as compared to the controls. There was no significant difference in the levels of serum FT4 and Serum TSH.

Key Words: Serum FT3, Serum FT4, Serum TSH, Oxidative stress and antioxidants

INTRODUCTION

Diabetes mellitus is the most common endocrine metabolic disorder, affecting about 170 million people worldwide [1]. The diseases of the thyroid gland are amongst the most abundant endocrine disorders in the world which are second only to diabetes. Thyroid diseases affect approximately 10-15% of the patients with diabetes, whereas in non-diabetics, the prevalence is approximately 6%. The mode of association between diabetes and thyroid diseases is more complex and is largely unclear. Neither diabetes nor thyroid diseases present a homogenous, nosological unit; the pathogenesis of the different types of diabetes as well as thyroid diseases is diverse. Therefore, even the correlations between them are different [2].

The metabolic dysregulation which is associated with diabetes mellitus causes secondary pathophysiological changes in multiple organs due to hyperglycaemia. Prolonged exposure to elevated glucose induces both repeated acute changes in intracellular metabolism and cumulative long-term changes in the structure and function of the macromolecules [3]. In a normal cell, there is an appropriate pro-oxidant- antioxidant balance. This balance is shifted towards pro-oxidants when the production of the oxygen species is increased or the levels of the antioxidants are decreased and this state is called as 'oxidative stress'. Oxidative stress is implicated in the pathogenesis of a variety of human diseases [4].Oxidative damage occurs to biomolecules like lipids, proteins, carbohydrates and nucleic acids and other extracellular components like collagen and hyaluronic acid which are very deleterious [5].The formation of lipid peroxides by the action of the free radicals on unsaturated fatty acids has been implicated in the pathogenesis of atherosclerosis and vascular diseases [6].Increased levels of the products of oxidative damage to lipids have been detected in the sera of diabetic patients and their presence correlates with the development of complications [7]. The body's defense mechanisms play an important role in the form of antioxidants that help to minimize the damages which are caused by oxidative stress. Antioxidants are compounds that dispose, scavenge and suppress the formation of free radicals or oppose their actions [8]. Hence, the present study was undertaken to assess the thyroid hormone status, the extent of lipid peroxidation and the status of the antioxidant defense mechanisms in patients with diabetes.

MATERIALS AND METHODS

The present study was conducted in the Department of Biochemistry, PES Medical College, Kuppam. Thirty diagnosed cases of type-2 diabetes mellitus were chosen for the study, which belonged to the age-group of 35-60 years. None of them had a previous history of thyroid diseases. Thirty age-matched subjects without diabetes were taken as the controls. Informed consent was obtained from all of them. Ten milliliters of fasting blood samples were collected by venipuncture and for the separation of sera, 5 ml of blood was centrifuged at 3000rpm for 5min and the remaining 5ml of blood was taken into a plain vial containing EDTA and was centrifuged at 3000rpm for 10min for the separation of plasma. The separated serum was used to estimate Serum TSH, FT3 and FT4 by the ELISA method [9], [10]. The plasma MDA levels were estimated by using thiobarbituric acid reacting substances (TBARS) by the method of Yagi [11] and Sinnhuber et al [12]. Reduced glutathione was determined by the method of Beutler et al [13] and ascorbic acid was determined by the method of Tietz [14]. All the results were expressed as mean ±SD and statistical comparsions were done. Due permission was obtained from the Institutional Ethics Committee prior to the starting of the work.

RESULTS

[Table/Fig 1]

	Type-2 Diabetics(n=30) Mean±S.D	Controls(n=30) Mean±S.D	p value
FT3(pmol/l)	3.11±0.52	3.76±0.57	< 0.05 (significant)
FT4(pmol/l)	9.33±0.26	9.26±0.28	Not significant
TSH(µIU/ml)	3.51±0.55	3.58±0.49	Not significant

[Table/Fig 1]: Comparison of FT3, FT4 and TSH levels in type-2 diabetics with controls.

[Table/Fig 2]

	Type-2 diabetics(n=30) Mean±S.D	Controls(n=30) Mean±S.D	p value

MDA (nmol/ml)	6.98±0.13	2.41±0.12	< 0.001 (highly significant)
GSH (mg/g Hb)	8.96±0.21	14.76±0.13	< 0.001 (highly significant)
Vit-C (mg/dl)	0.61±0.17	2.13±0.19	< 0.001 (highly significant)

[Table/Fig 2]: Comparison of MDA, GSH and vitamin-C levels in type-2 diabetics with controls.

There was a significant decrease in the values of serum FT3 in type-2 diabetics as compared to the controls. There was no statistically significant difference in the values of serum FT4 and TSH between the type-2 diabetics and the control groups [Table/Fig 1]. There was a significant increase in the values of MDA in diabetes mellitus patients as compared to the controls, whereas, there were significantly decreased levels of Vit-C and reduced glutathione in the diabetes mellitus cases as compared to the controls [Table/Fig 2].

DISCUSSION

Diabetes mellitus comprises a group of common metabolic disorders that share the phenotype of hyperglycaemia. Several distinct types of diabetes mellitus exist and these are caused by a complex interaction between the genetic factors [15].In the present study; there was no statistically significant difference in the levels of serum FT4 and serum TSH. The decreased serum level of FT3 may be due to the impairment of the 5-monodeiodinase enzyme activity which controls the peripheral conversion of T4 into T3 [16].Suzuki et al [17] attributed the abnormal thyroid hormone levels which are found in diabetes to the presence of the thyroid hormone binding inhibitor (THBI), which is an inhibitor of the extra thyroidal conversion enzyme which converts T4 to T3 and to the dysfunction of hypothalamo-hypophyseal-thyroid axis. These situations may prevail in diabetics and would be aggravated in poorly controlled diabetics. Stress, when associated with diabetes, may also cause changes in the hypothalamo anterior-pituitary axis. It appears that sub-clinical hypothyroidism and hyperthyroidism may result from the hypothalamo-hypophyseal-thyroid axis disorders, as suggested by Celani et al [18]. A suggestion was made that the finding of definite hypothyroidism or hyperthyroidism should be given adequate attention and that the treatment of the thyroid disorder should be appropriately undertaken [19].

The significant rise in the MDA levels in diabetes confirms that it is associated with an increased production of reactive oxygen species (ROS) and free radicals. Lipid peroxidation is a chain reaction which provides a continuous supply of free radicals that initiate further peroxidation [20]. Insulin secretion is associated with peroxide production. We also observed a significant decrease in the levels of reduced glutathione in the cases as compared to the controls. The intracellular depletion of reduced glutathione (GSH) can be either due to the formation of a direct complex with an electrophilic agent or due to the inhibition of synthesis or due to the subjection of the cell to oxidative stress [21]. When a cell is subjected to oxidative stress, there is increased utilization of glutathione, thus leading to its depletion. Many enzymes are GSH dependent and their activity may be regulated by the thiol disulphide exchange. They are thus dependent on the GSH status. Glutathione-S-transferase (GST) is reduced in diabetics which are dimeric, mainly cytosolic enzymes that have extensive ligand binding properties in addition to their catalytic role in detoxification [22], [23]. This reduction is due to the reduced levels of GSH. There is also a decrease in the levels of non-enzymatic anti-oxidants such as Vit-C, which states that there is an increased defense mechanism against oxidative damage in diabetes mellitus. The decrease in the levels of these non-enzymatic antioxidant parameters may be due to an increased turnover for preventing oxidative damage in these patients, thus suggesting an increased defense against oxidative damage [24].

It could be concluded that type-2 diabetic patients are more prone to thyroid abnormalities, which requires regular follow-up. Oxidative stress may play a major role in the pathogenesis and the development of diabetes mellitus and a decreased antioxidant status shows a necessity for the therapeutic administration of antioxidants as supplementary therapy to the diabetic cases. Extensive studies are required in future to reduce the morbidity of diabetes mellitus and its complications.

REFERENCES:

[1] Wild S, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes. Estimates for the year 2000 and projections for2030.Diabetes Care, 2004; 27:1047-1053.
[2] Perros P, Mc crimmon RS, Shaw G, Frier BM. Frequency of thyroid dysfunction in diabetic patients: value of annual screening. Diabetes Med, 1995:12(7):622-627.
[3] Sheetz MJ, King GL. Molecular understanding of hyperglycemia's adverse effects for diabetic complications. JAMA, 2002; 288:2579-2588.
[4] Beck MM, Levander OA. Dietary oxidative stress and potentiation of viral infection. Annu Rev Nutr,1998; 18:93-116.
[5] Frei B. Reactive oxygen species and antioxidant vitamins: mechanisms of action. Am J Med, 1994; 97: S5-S13.
[6] Donald DH. Oxidative stress and vascular disease, Arterioscler Thromb Vasc Biol 2005; 26:689-695.
[7] Liu SX, Hou FF, Guo ZJ et al: Advanced oxidation protein products accelerate atherosclerosis through promoting oxidative stress and inflammation. Arterioscler Thromb Vasc Biol, 2006; 26:1156-1162.
[8] Krishna Mohan S and Venkata Ramana G. Status of lipid peroxidation, glutathione, ascorbic acid, and vitamin E and antioxidant enzymes in patients with pregnancy induced – hypertension (PIH). Indian J Physiol Pharmacol, 2007; 51 (3): 284 –288
[9] Frazer CG, Browing MCK. Measuring serum thyroglobulin. Lancet 1985; 816-819.
[10] Holl RW, Bohm B, Loos U et al. Thyroid autoimmunity in children and adolescents with type-1 DM. Effects of age, gender and HLA type. Horm Res,1999; 52(3).113-118.
[11] Yagi K. Lipid peroxides and human diseases. Chem Phys Lipids, 1978; 45: 337-351.
[12] Sinnhuber RO, Yu TC, Yu TC. Characterization of red pigment formed in thiobarbituric acid determination of oxidative rancidity. Food Res, 1958; 23: 626-630.
[13] Beutler E, Duron O, Kelly BM. Improved method for determination of blood glutathione. J Lab Clin Med, 1963; 61: 882-888.
[14] Tietz NW (Ed.).Textbook of Clinical Chemistry. W.B. Saunders Company, Philadelphia, London, Toronto.2004; pp.960-962.
[15] Kasper DL, Braunwald E, Hauser S, Longo D, Jameson JL, Fauci AS(Eds.).Diabetes mellitus. In: Harrison's Principles of Internal Medicine.16th Edn. 2004. McGraw Hill Professional, USA. pp. 2152-2180.
[16] Suzuki Y: Nanno M: Gemma R et al. Plasma free fatty acids inhibitor of extra thyroidal conversion of T4 to T3 & thyroid binding inhibitor in patients with various non-thyroidal illnesses. Endocrinol Jpn, 1992; 39:445-53.
[17] Suzuki J, Nanno M, Gemma R, Tanaka I, Taminato T, Yoshimi T. The mechanism of thyroid abnormalities in patients with diabetes mellitus. Nippon Niabunpi Gakki Zashi 1994; 7(4):465-70.
[18] Celani HF, Bonati HE, Stucci N. Prevalence of abnormal thyrotropin concentrations measured by a sensitive assay in patients with type 2 diabetes mellitus. Diabetes Res, 1994; 27 (1) 15-25.
[19] Suzuki H, Hiraiwa M, Suzika Y. Hasigam Y, Shimoda S. Thyroid function in non-thyroidal illness. Specific changes in serum levels of thyroid hormones in patients with non-thyroidal illness. Nippon Niabunpi Gakki Zashi, 1984; 60 (6) 738-55.
[20] Murray RK, Granner DK, Rodwell VW (Eds.).Harper's Illustrated Biochemistry. 27thEdn.2006.McGraw Hill Lange International Edition. pp.128-129.
[21] Deneke SM, Farberg TZ. Regulation of cellular glutathione lung cells Mol.Physiol,1994; 1163-1173.
[22] Listowsky I, Abramovitz M, Homma H et al. Intracellular binding and transport of hormones and xenobiotics by glutathione-s-transferase. Drug Metab Res, 1988; 19:305-318.
[23] Ketley JN, Habig WH, Jacoby WB. Binding of non substrate ligands to glutathione-s-transferases. J Biol Chem, 1976; 250:8670-8673.
[24] K M Surapaneni , Venkata Ramana G. Status of lipid peroxidation, glutathione, ascorbic acid, vitamin E and antioxidant enzymes in patients with osteoarthritis. Indian J Med Sciences, 2007; 61 (1): 9 – 14

AUTHORS:
1. KEDARI G S R

NAME OF DEPARTMENT(S) / INSTITUTION(S) TO WHICH THE WORK IS ATTRIBUTED:
Dept of Biochemistry, Saveetha Medical College & Hospital, Saveetha University, Saveetha Nagar, Thandalam, Chennai – 602 105, Tamilnadu, India.

NAME, ADDRESS, PHONE, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Kedari G S R, Assistant Professor, Dept of Biochemistry, Saveetha Medical College & Hospital, Saveetha University, Saveetha Nagar, Thandalam, Chennai – 602 105, Tamilnadu, India.
Email: kedari.gsr@gmail.com

DECLARATION ON COMPETING INTERESTS: No competing Interests

Date of Submission: **Jan 31, 2011**
Peer Review Completion: **Feb 21, 2011**
Date of Acceptance: **Mar 04, 2011**
Date of Publication: **Apr 11, 2011**