

# Analysis of Level of Antioxidants and Oxidative Stress in Diabetic Patients

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## ABSTRACT

**Background and objectives:** Diabetes mellitus is a metabolic disorder whose prevalence is rising rapidly in an alarming rate. The main objective of the study is to analyse the level of antioxidants and oxidative stress in diabetic patients among local population of Pakistan. **Material and methods:** This cross sectional study was conducted in Department of Pathology, Islamic International Medical College Rawalpindi during March 2019 to November 2019. For this purpose we selected the patients who were suffering from diabetes. The data was collected from 100 patients who visited the OPD of the hospital. This study is based on the local population of Pakistan, which shows the stress level in diabetic patients.

**Results:** The data was collected from 100 patients. According to analysis of data level of antioxidant and oxidative stress is increasing in diabetic patients because

cell becomes destroyed. GSH is important non-enzymatic antioxidant which helps in scavenging of free radical mechanism. According to data the levels of GSH become decreases in diabetic patients. The data pertaining in the table shows that levels of sialic acid become increases in patients.

**Conclusion:** It is concluded that level of antioxidants in our body plays an important role. It is obvious from the presented data that a relation exists between hyperglycaemia, oxidative stress, cellular and endothelial dysfunction.

**Key words:** Antioxidants, Control, Enzymes, Radical, Diabetes

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## INTRODUCTION

Diabetes mellitus is a metabolic disorder whose prevalence is rising rapidly in an alarming rate. This disease has shown a change in its condition over the last 30 years from a mild disorder of the elderly to the major cause for morbidity and mortality affecting the youth and middle-aged people. India leads the world with the largest number of diabetic patients (Cosentino F *et al.*, 1998), earning the distinction, the “diabetes capital of the world.” Although there is an increase in the prevalence of type I diabetes, the most common form of diabetes is type II, which accounts for more than 90% of all diabetes cases (Guigliano D *et al.*, 1996). Diabetes mellitus is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion and insulin action or both (Kesavulu M *et al.*, 2000).

The chronic hyperglycemia is associated with long-term damage, dysfunction, and failure of normal functioning of various organs, especially the eyes, kidneys, nerves, heart, and blood vessels. Diabetes-specific micro vascular disease is a leading cause of blindness, renal failure, and nerve damage (King G *et al.*, 1994). Diabetes is a major source of morbidity, mortality, and economic cost to the society. People with diabetes showed the risk of the development of acute metabolic complications such as diabetic ketoacidosis, hyperglycaemic hyperosmolar non-ketotic coma, and hypoglycaemia (Koya D *et al.*, 1998).

Oxidative stress is caused by an unfavourable balance between Reactive Oxygen Species (ROS) and antioxidant defences. ROS are generated during normal cellular metabolism, as a result of the influence of various environmental factors, as well as during pathological processes. Reactive oxygen species play an important role in the pathogenesis of cancer<sup>1</sup>. Oxidative stress caused by increased free radical generation and/or decreased antioxidant level in the target cells and tissues has been suggested to play an important role in carcinogenesis (Ruderman

N *et al.*, 1992).

Free radicals are capable of altering all major classes of biomolecules, such as lipids, nucleic acids and proteins, with changes in their structure and function. Prime targets of free radicals are the polyunsaturated fatty acids in cell membranes and their interaction results in lipid peroxidation (Miata D *et al.*, 2000). The levels of free radical molecules are controlled by various cellular defense mechanisms, consisting of enzymatic (catalase, glutathione peroxidase, superoxide dismutase) and non-enzymatic (vit. E, vit. C, glutathione) components (Ho E *et al.*, 1999).

The main objective of the study is to analyse the level of antioxidants and oxidative stress in diabetic patients among local population of Pakistan.

## MATERIALS AND METHODS

This cross sectional study was conducted in Department of Pathology, Islamic International Medical College Rawalpindi during March 2019 to November 2019. For this purpose we selected the patients who were suffering from diabetes. The data was collected from 100 patients who visited the OPD of the hospital. This study is based on the local population of Pakistan, which shows the stress level in diabetic patients.

### Inclusion criteria

- All those patients who confirmed diabetes mellitus by fasting blood sugar were included in this study.

### Exclusion criteria

- Patients with, habits of smoking, alcoholism, other systemic diseases such as coronary artery disease and renal diseases were excluded in this study.

### Data collection

5.0 ml blood sample was taken from vein. Blood was further processed for the estimation of GSH, Catalases, SOD, MDA, Neuramin-

idase and Sialic acid. Commercially available enzymatic kits of Randox were used. Blood was centrifuged at 4000 rpm for 10 minutes and serum was separated. Blood samples will be collected into EDTA tubes from fasting proteins. The blood will be centrifuged and indomethacin and butylated hydroxytoluene will be added into the plasma samples before they will be stored at -80 °C until analysis. The sample were processed and analysed for the estimation of SOD, GSH, CATALASES, MDA, NO, neuraminidase and Sialic acid levels.

**Statistical analysis**

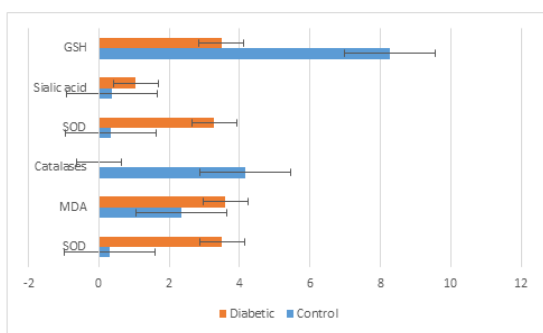
The collected data were analysed using SPSS software (version 19). The results are presented as a mean with 95% confidence interval limits or standard deviations. The significant value for P<0.05 was accepted as statistically significant.

**RESULTS**

The data was collected from 100 patients. According to analysis of data level of antioxidant and oxidative stress is increasing in diabetic patients because cells become destroyed. GSH is important non-enzymatic antioxidant which helps in scavenging of free radical mechanism. According to data the levels of GSH become decreases in diabetic patients. The data pertaining in the table shows that levels of sialic acid become increases in patients. The level becomes increases in all cases. As the value in this case is 3.48 ± 0.65. According to our data MDA is considered to be an important antioxidant and serum stress biomarker in case of diabetic patients (Table 1 and Figure 1).

**Table 1: Level of anti-oxidants in control and diabetic patients**

Variable	CONTROL (moles/ml)	(moles/ml)
		(n=100)
SOD	0.32	3.5 ± 0.74
MDA	2.35	3.6 ± 0.82
Catalases	4.16	0.00 ± 0.00
SOD	0.326	3.27 ± 0.16
Sialic acid	0.37	1.05 ± 0.08
GSH	8.26	3.48 ± 0.65
Mean ± SD		



**Figure 1: Graphical view of level of anti-oxidants in control and diabetic patients**

**DISCUSSION**

Reactive Oxygen Species (ROS) cause oxidation of DNA, proteins and lipids, and induce carcinogenesis. Some studies have reported high lipid peroxidation levels become high in human colorectal cancer tissue and gastric cancer tissue (Ho E *et al.*, 1999). The major aldehyde products of lipid peroxidation are Malondial-Dehyde (MDA) and 4-hydroxynonal. MDA is mutagenic in mammalian cells and carcinogenic (Hunt J *et al.*, 1988).

Peroxidation of lipids can disturb the assembly of the membrane, causing changes in fluidity and permeability, alterations of ion transport and inhibition of metabolic processes. Injure to mitochondria induced by lipid peroxidation can direct to further ROS generation (Schmidt K *et al.*, 1995). Catalase is a common enzyme found in nearly all living organisms which are exposed to oxygen, where it functions to catalyze the decomposition of hydrogen peroxide to water and oxygen. Catalase has one of the highest turnover numbers of all enzymes; one molecule of catalase can convert millions of molecules of hydrogen peroxide to water and oxygen per second (Arora R *et al.*, 2013).

Superoxide is one of the main reactive oxygen species in the cell and as such, Super Oxide Dismutase (SOD) serves a key antioxidant role. The physiological importance of SODs is explained by the severe pathologies evident in mice genetically engineered to lack these enzymes (Halliwell B *et al.*, 1993). In mammals there are several types of SODs, which differ with respect to their location in the cell and the metal ion they require for their function. For example, a copper-zinc SOD is present in the fluid filling the cell (i.e., the cytosol) and in the space between two membranes surrounding the mitochondria (Diplock AT *et al.*, 1998). Furthermore, a manganese-containing SOD is present in the mitochondrial interior. Both of these enzymes are critical for prevention of ROS-induced toxicity (Kakkar P *et al.*, 1984).

The increase in the lipid peroxidation status and the decline in the antioxidant defense mechanism are the prime factors in the development of complications in diabetics. These biochemical parameters can be evaluated from the serum samples. Saliva values of the same are much less; nevertheless, it can be estimated from this body fluid too. In our study, we estimated the oxidative stress status by measuring MDA and the antioxidant status by measuring GSH and SOD in the serum and saliva of age- and sex-matched diabetics and non-diabetics (Mahadevan K *et al.*, 2012). Our result could not only substantiate the diagnostic potential of saliva as a body fluid, but also predict the serum values of MDA and GSH from the corresponding saliva values in diabetics and non-diabetics (Arana C *et al.*, 2006).

**CONCLUSION**

It is concluded that level of antioxidants in our body plays an important role. It is obvious from the presented data that a relation exists between hyperglycaemia, oxidative stress, cellular and endothelial dysfunction.

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