Serum Levels of Protein Carbonyl and Electrolytes in Patients with Type 1 Diabetes Mellitus

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Abstract Type 1 diabetes mellitus (T1DM) is a disease characterized by the insufficient insulin in a genetically prepared people, and mostly appeared at young ages. Therefore, T1DM is a chronic metabolic disease, and like other metabolic diseases, it associates with elevated oxidative state. Reactive oxygen species (ROS) are metabolites with high reactivity that can cause a diverse destruction to the cellular compartments. Proteins are clear targets for ROS, in which yields oxidized proteins, including protein carbonyls. In the present study, our goal was to determine the oxidative status of T1DM patients by using protein carbonyls as a biomarker. The relationship of protein carbonyl with serum electrolytes (Na^+, and K^+) were investigated as well. The study was included 60 adolescent with T1DM disease, and 30 healthy adolescent as control. The level of protein carbonyl was elevated significantly in the serum of T1DM patients (24.17±5.93 ng/mL) compared to the control adolescent (12.77±3.11 ng/mL). On the other hand, the levels of both Na and K were reduced in the serum of T1DM patients. The level of oxidative stress was reached the oxidative damage of proteins systemically in these patients. This may results in increasing the health risks of diabetes and may influence the proper growth of these adolescents. No association was observed between protein carbonyl and electrolytes in T1DM patients, but a significant association was obtained between glucose levels and Na levels. We suggest the administration of antioxidants with glycemic control in T1DM patients.

Keywords: T1DM, glucose, PCO, Na^+, K^+.

1. Introduction

Type 1 diabetes mellitus (T1DM) is assumed to be caused via the death of insulin producing pancreatic cells that is immune associated, if not directly immune mediated [1, 2]. T1DM was once thought to be primarily a condition of children as well as adolescents, but this perception has shifted in the last decade, as well as age at symptomatic start is no longer a limiting factor [3]. In children as well as also adolescents, polyphagia, as well as to a lesser extent in adults, polydipsia, as well as polyuria (the classic trio of symptoms associated with disease start) remain diagnostic markers, coupled with overt hyperglycemia. T1DM is also

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defined by the necessity for exogenous insulin replacement right away, necessitating lifelong management. T1DM prevalence, pharmaceutical efficacy, understanding how the disease develops, as well as preventing or treating the disease are all unanswered questions [4].
T1DM is growing in both incidence as well as prevalence over the world, with annual increases in incidence of roughly 2–3% per year [5, 6]. Retinopathy, neuropathy, as well as nephropathy are the more common microvascular complications of the disease, although they can also impact cognitive function, the heart, as well as other organs. Hyperglycemia is the key risk factor for microvascular disease, and lowering glycated hemoglobin through rigorous diabetes care, especially early in the disease, is related to dramatic (70 percent) reductions in microvascular disease incidence as well as progression [7].
Metabolic diseases are associated with the progression of oxidative damage owing to the increase the production of reactive oxygen species (ROS) [8, 9]. These reactive metabolites are normally perform a vital cellular function [10], but when their level exceeds the physiological condition, it would lead to cellular destruction because of their reactivity [11, 12]. Essentially, the ROS are detoxified by a cellular substances called antioxidants, which comprise a small molecules such as coenzyme Q10, reduced glutathione, and uric acid, and large molecules such as the enzymatic antioxidants [13]. Therefore, ROS can react with the macromolecules of the cells, including proteins, fatty acids, and nucleic acids [14-16]. This oxidative damage appeared only when the antioxidant substances cannot detoxify all of the ROS in the cells [17]. The protein oxidation caused by the damaging effect of ROS [18, 19] can be monitored by measuring the protein carbonyl levels [20]. This imbalance between ROS and antioxidants is termed as oxidative stress [21]. This study was took the fact that T1DM is a metabolic disease, and hence, we aimed to measure the oxidative damage level, by determining the protein carbonyls in the serum of T1DM patients. The relationship of protein carbonyl with serum electrolytes (Na^+, and K^+) were investigated as well.

2. Materials and Methods

2.1. Patients

The patients were documented in the consultancy of Al-Yarmook Teaching Medical Hospital (Baghdad, Iraq). They were informed about the standard criteria of the research and agreed to become a volunteers in this work. 60 patients with T1DM were selected for the study from Jun to October 2021, and controlled with 30 healthy volunteered people.

2.2. Methods

The T1DM patients and healthy control people were donated a vein blood upon 8 hours of fasting. Then the blood was centrifuged in a medical centrifuge (4000 rpm for 10 minutes), as well as the serum was stored in a deep freezer at -20 °C to be analyzed for glucose and sodium, and potassium by using a spectrophotometric method (Apel PD-303, Japan) from commercial kits purchased from the agent of BIOLABO, France. The protein carbonyl concentration was determined by using an enzyme linked immune sorbent assay (ELISA) kit based on sandwich technology and the measurement was applied in ELISA microplate reader (Human, Germany).

2.3. Statistics

The data were processed statistically on the computer by a program from IBM called SPSS version 26.0, for mean comparisons in an independent sample t-test, and the relationship between glucose, Na, K, and protein carbonyl were calculated according to the Pearson correlation. At last, the sensitivity of protein carbonyl as diagnostic marker for T1DM was

determined by the receiver operating characteristic (ROC) curve through measuring area under curve (AUC) of each variable.

3. Results

The characteristics of the volunteered T1DM patients and control adolescent are contained in Table 1. Age was shown non-significant (P>0.05) differences between the T1DM patients (15.48±0.97 year) and control people (15.80±1.03 year). The levels of protein carbonyl were elevated significantly (P<0.05) in the serum of T1DM patient (24.17±5.93 ng/mL) compared to the control adolescent (12.77±3.11 ng/mL).

Table 1: Volunteered people characteristics.

Parameter	**T1DM**	**Control**	***P*-value**
N	60	30	-
Age (year)	15.80±1.03	15.48±0.97	0.166
Protein carbonyl (ng/mL)	12.77±3.11	24.17±5.93	0.0001
Glucose (mg/dL)	86.67±5.59	146.99±15.02	0.0001
Na (mEq/L)	142.47±1.41	139.86±2.04	0.0001
K (mEq/L)	4.52±0.26	4.20±0.31	0.0001

The level of glucose was observed to be significantly (P<0.05) higher in serum of T1DM patients (146.99±15.02 mg/dL) compared to the serum of control adolescent (86.67±5.59 mg/dL). Furthermore, Na levels was also observed to be significantly (P<0.05) reduced in the serum of T1DM patients (139.86±2.04 mEq/L) compared to the serum of control adolescent (142.47±1.41 mEq/L). Also, K level was reduced significantly (P<0.05) in the serum of T1DM patients (4.20±0.31 mEq/L) compared to the serum of control adolescent (4.52±0.26 mEq/L).
The results have shown significant negative association between glucose and Na in the serum of T1DM patients, as shown in Table 2.

Table 2: Correlation in T1DM patients.

Parameter	**Protein carbonyl (ng/mL)**		**Glucose (mg/dL)**		**Na (mEq/L)**		**K (mEq/L)**	
	r	*p*-value	r	*p*-value	r	*p*-value	r	*p*-value
Glucose (mg/dL)	-0.136	0.300	-	-	-0.837	0.0001	-0.092	0.485
Na (mEq/L)	0.113	0.391	-0.837	0.0001	-	-	-0.015	0.911
K (mEq/L)	-0.035	0.792	-0.092	0.485	-0.015	0.911	-	-
Age (year)	-0.083	0.530	0.139	0.289	-0.168	0.200	0.219	0.092

The ROC curve of protein carbonyl has indicated the usefulness of this biomarker in the diagnosis of T1DM disease. MDA has shown excellent sensitivity (AUC=0.974, $P<0.0001$) in the diagnosis of T1DM patients comparing to the healthy control adolescent, as shown in Figure 1.

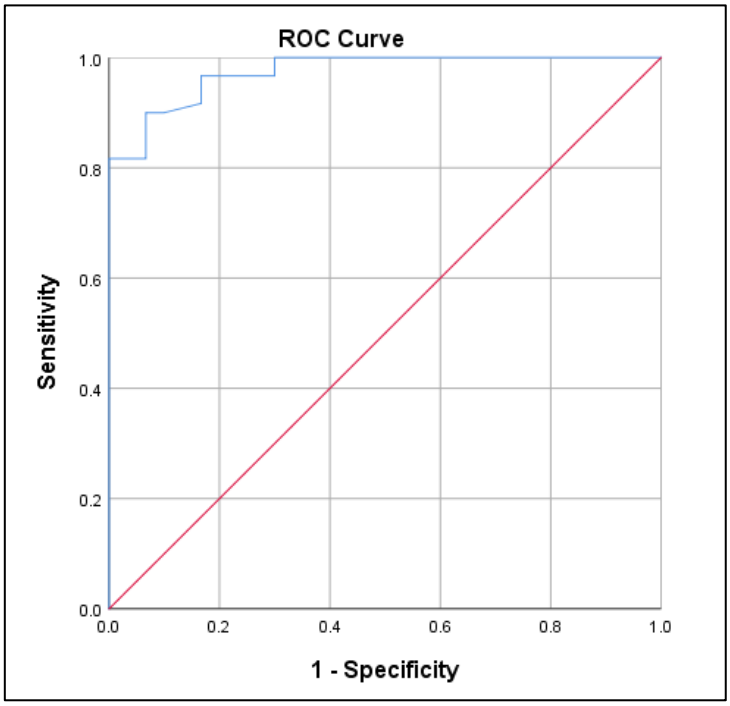

Figure 1: The ROC curve in the diagnosis of T1DM by using protein carbonyl as biomarker.

4. Discussion

Oxidative stress was reported in both T1DM [22] and T2DM diseases [23]. Telci *et al.* have reported significant increase in the levels of protein carbonyl in T1DM patients compared to their control. They have confirmed a redox imbalance in T1DM results in the appearance of oxidative stress, and they have attribute this imbalance to metal-catalyzed protein oxidation in T1DM patients clinically free from complications [24].

Dominguez *et al.* have indicated an increase in the plasma lipid peroxidation and protein carbonylation in children and adolescent with T1DM disease. The authors have concluded that systemic oxidative stress is present upon early onset of T1DM as well as is increased via early adulthood [25]. In our study, the increase of protein carbonyl level was highly significant in T1DM adolescent, but it does not associated with the serum glucose levels.

The levels of both Na and K were reduced significantly in T1DM patients. Al-Rubeaan *et al.* have reported similar results, in which they have found a negative correlation between Na and glucose, but no association of glucose to the reduction of K [26], which was agreed with the present observations. Datchinamoorthi and Rajagopalan have reported that the link between blood glucose as well as serum electrolytes is multi factorial in which it is linked to a number of other factors, which includes age as well as linked conditions [27]. High glucose levels raise serum osmolality, causing water to flow out of the cells and, as a result, a fall in serum Na levels by dilution [28].

5. Conclusions

The results were revealed an elevated oxidation state in the serum of T1DM patients. The levels of oxidative stress were reached the oxidative damage of proteins systemically in these patients. This may results in increasing the health risks of diabetes and may influence the proper growth of these adolescents. No association was observed between protein carbonyl and electrolytes in T1DM patients, but a significant association was obtained between glucose levels and Na levels. We suggest the administration of antioxidants with glycemic control in T1DM patients.

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