Measurement of Trace Elements Association with Diabetes Mellitus Based on Atomic Absorption Spectrophotometers

Dr. Auns Q. Hashim Al-Neami
Ministry of Higher Education and Scientific Research
Al-Nahrain University / College of Engineering/ Medical Eng. Dep.
E-mail: uns_alneami@yahoo.com

Abstract
The aim of the present study was to investigate the serum level of zinc (Zn), chromium (Cr), and magnesium (Mg) in random population of diabetic mellitus patients with type 1 and 2 diabetes mellitus and their possible association with age, and sex. A relationship has been reported between trace elements and diabetes mellitus. The trace elements were very important for human life, which their equilibrium is healthy and their imbalance in blood serum lead to very dangerous diseases. This study evaluated the role of such a relationship between the plasma zinc, chromium and magnesium and diabetes mellitus. (Zn, Mg, and Cr) have been measured in the serum of in 20 patients (12 men and 8 women) apparently healthy Iraqi persons aged (20-70) years. The levels of these elements were analysed using flame and flameless atomic absorption spectrophotometer, Phoenix-986 AA spectrophotometer and Shimaadzu AA-6800 atomic absorption spectrophotometer respectively. The mean concentration of zinc was between (0.76 – 1.02) µg/ml in men and slightly lower in women, it was in the range (0.71 – 1.01) µg/ml. The concentration of zinc were slightly increased with ages especially the groups between (30-42) years and slightly reduced in the group between (49-60) years. The concentration of zinc in all groups under investigation seems to be stable. The study shows that there is a deficiency in zinc value in men and women controls by 23.5% and 20.1% respectively. The chromium level of blood serum were measured for all volunteer, the mean range in men were between (0.012 – 0.060) µg/ml with a deficiency of 40.1%. While the chromium range in women was between (0.011 – 0.041) µg/ml with a deficiency of 40.4%. The result observed that the concentration of chromium has the highest value in both sexes were in the age of (23-32) years. The mean concentration of magnesium in blood serum was measured for all adults under investigation. It was between (18 – 27) µg/ml for men and women. It was noted that the Mg level was more in women than in men in the ages between (50-59) years. The mean Mg deficiency was 12.3%. The goal of this study was to know the effect of trace element especially (Zn, Mg, and Cr) on the diabetes mellitus by measuring the concentrations of these elements and determine the deficiency of each trace element in the patients and then treat the state.

Keywords: Trace elements, Diabetes mellitus, Zinc, Chromium, Magnesium

1. Introduction
Diabetes Mellitus is the commonest major metabolic disease and most prevalent diseases worldwide. Its related morbidity is due to its micro and macro angiopathic complications [1]. Trace elements are tiny molecules that play important roles in our bodies; their metabolism has been reported to alter in diabetes mellitus and might have specific roles in the pathogenesis and progress of this disease [2]. The growing concern with environmental factors in human health over the last few years has aroused renewed interest in the trace elements. Abnormalities in their metabolism have been demonstrated in many human diseases. In particular, diabetes mellitus has been shown to be associated with abnormalities in the metabolism of zinc, chromium, magnesium [3].

The literature review shows that a relationship was observed between diabetes mellitus and trace elements in many research studies. Candilish [4] trace elements as (Cu, Fe, and Se) play important role in insulin action including activation of insulin receptor, serving as cofactor or components for enzyme systems involved in glucose metabolism. Bushra F. H.[5] investigate the serum level of copper (Cu), zinc (Zn), selenium (Se), iron (Fe) in women with type 2 diabetes mellitus and their possible association with lipid profile.

Trace elements are widely distributed in nature in variable proportions according to geographical sites. Which lead to variation food intake of these elements according to their availability, and they play a vital role in growth, health, and maintenance of human body in the same way as the protein, vitamin and other essential nutrients do [6]. The most dominant feature of the metabolism in diabetes is an abnormally high concentration of blood glucose. Trace elements of great important for their therapeutic efficacy, and their lack may result in characteristic pathological sign and symptom, therefore, enough daily intake should not lead to any disease disturbances [7], on other hand, it may be toxic in higher concentrations. These elements are especially prominent in assisting enzyme functions, and few of them act as biological catalysts and coenzymes [8]. The clinical interest in trace element determination, for diagnosis and prognosis of different diseases related to deficiency or toxicity of these elements [9], for examples:

**Zinc**: Zinc (Zn) is the most abundant trace element in the human body and is an important nutrient and cofactor of numerous enzymes and transcription factors that is involved in various physiological metabolisms. Zn
dyshomeostasis is often associated with various pathogeneses of chronic diseases, such as metabolic syndrome, diabetes, and related complications[10]. Scott and Fischer first recognized the relationship between zinc and insulin. They found that whereas the normal human pancreas contained significant quantities of zinc, the diabetic pancreas contained very little. Later, the availability of histochemical techniques for the detection of zinc confirmed that zinc and insulin concentrations in the pancreas changed in the same direction in a variety of situations in humans [11]. Organic compounds which were capable of reducing the zinc content of the pancreas were found to be diabetogenic in animal experiments [12]. Meltzer et al appear to be the first to have studied the urinary excretion of trace elements in diabetes mellitus. As a group, diabetics excrete more zinc in the urine than non-diabetics [13]. The mean plasma, leucocyte and erythrocyte zinc levels are significantly lower in diabetics than in non-diabetics. This has been challenged by other workers. No correlation has been found between the plasma or urinary zinc levels on the one hand and the age at onset of diabetes, the patient's age or his weight, on the other [14].

The function of zinc in the body metabolism is based on its enzymatic affinity and way of a zinc enzyme complex or metallo-enzyme. In humans and animals, diabetes causes disturbances in this vital trace element. Zinc is required for insulin synthesis and storage and insulin is secreted as zinc crystals. It maintains the structural integrity of insulin [15]. Zinc has an important role in modulating the immune system and its dysfunction in diabetes mellitus may be related in part to the status of zinc [16]. Therefore; serum levels of zinc are usually found low in diabetic patients. Zinc affects the antigenic properties of insulin and the binding of insulin to hepatocyte membranes and a deficiency can lead to increased insulin resistance and hyperglycemia [17]. Elevated glucose in turn produces hyperzincuria. Low zinc leads to poor or slowed wound-healing common in diabetic patients. Zinc is required for insulin storage and cellular binding, although high concentrations can lead to a reduction in insulin release [18].

Chromium: The biological activity of chromium (Cr) depends on its valency and the chemical form of the complex of which it is a part. Glucose tolerance factor (GTF) is a trivalent form of chromium that has high biological activity. This is required for optimal glucose utilisation by the cells [19]. The abnormalities in chromium metabolism exist in insulin dependent diabetics seems certain. It has been suggested that by acting on the ribosomes chromium facilitates the insulin stimulated aminoacid incorporation into protein. The glucose intolerance seen as age advances has been attributed to chromium deficiency [20], if chromium administration can be shown to consistently correct the glucose intolerance of ageing; it might replace the use of the less physiological oral hypoglycemic agents in the treatment of this condition. It is a cofactor for the initiation of peripheral insulin action on the receptors on the cell membranes [21].

Insulin dependent diabetics excrete more chromium than the control subjects. However, there is no significant difference in the urinary excretion of chromium between maturity onset diabetics and normal controls [22]. Chromium deficiency has also been held responsible for vascular complications associated with diabetes mellitus. The chromium deficiency leads to impaired tolerance, elevation of circulating cholesterol, and aortic plaque. The Cr deficiency play a significantly role in diabetes and cardiovascular diseases. In such cases Cr supplementation becomes absolutely essential [23].

Magnesium: Magnesium (Mg) has an important role in the phosphorylation reactions of glucose and its metabolism. Its deficiency has been implicated in insulin resistance, carbohydrate intolerance, dislipidemia and complications of diabetes [24]. The association between diabetes mellitus and hypomagnesaemia is compelling, because of its wide ranging impact on diabetic control [25].

Magnesium is critical to the proper functioning of many physiological reactions, including those that are critical to the cardiovascular system. It is important in pathogenesis and treatment of cardiovascular disease [26]. However, magnesium abnormalities may be important in pathogenesis and magnesium replacement may be necessary for treatment is ischemic heart disease [27]. Magnesium deficiency can paradoxically increase the risk of, or protect against oncocogenesis over 300 enzymes that influence the metabolism of carbohydrate protein, nucleic acid, and ion transport require Mg [28]. So, Magnesium is known to be related to the carbohydrate and fat metabolism. Serum magnesium levels have been shown to be inversely related to the severity of diabetes [29]. Define lowering of serum magnesium has been shown in patients on long term treatment with insulin and those recovering from diabetic ketoacidosis [30]. Magnesium plays a role in the release of insulin, magnesium depletion has an atherogenic potential [31]. Hypomagnesaemia has been postulated as a possible risk factor in the development and progression of diabetic retinopathy [32].

2. Material and Methods:
Study population: The study population is known diabetic patients attending National Diabetes Center and Popular Clinic of Bab-Al Muathem in Baghdad.
Subject population: The present study included (20) patients categorized into (12 men and 8 female), and with
insulin and non-insulin dependent diabetes mellitus, their age ranged between (20-70) years.

**Instrumentation:** The comparative study included determination of trace elements using A Phoenix-986 AA flame spectrophotometer as in Fig.(1) and Shimaadzu AA-6800 flameless atomic absorption spectrophotometer as in Fig.(2).

**Blood sample collection:** Samples obtained for the study of normal level were obtained via plastic cannula in the vein. The blood was transferred to an acid-washed centrifuge tube and allowed to stand. After centrifugation, the serum was transferred to an acid-washed sample tube.

Sample pre-treated consists only of dilution (2X) directly into auto sampler cup. All water used to dilute standards and samples was deionized and distilled. Acids used were BDH “Analar” grade. Standard of elements were purchases from Aldrich 1000 ppm for each element used in this study. All glassware, pipettes, micropipettes tips and auto sampler cups were acid wash before use. Appropriate standards are prepared in 0.1 M nitric acid.

3. **Results**

Trace metals determination includes measurement the concentration of Cr, Zn, and Mg, the present instrumental setting during the tests is shown in table (1), and the characteristic of the Cr, Zn, and Mg tests are shown in the Fig.s (3), (4), and (5). In this study Matlab version 2013 program was used to determine the absorbance of the trace elements during the tests as shown in table (2).

4. **Conclusions**

Diabetes has become an international health care crisis that required new approaches to prevention and treatment. Diabetes management should begin with exercise and diet. Diabetes has been shown to be associated with abnormalities in the metabolism of Zn, Cr, and Mg. The impairment of these metals had been reported as aggravating factors in the progression of disease.

In order to understand the role and status of trace elements in diabetic patients, this study was undertaken to the serum zinc, chromium and magnesium levels in diabetic patients. Serum of these elements levels were found to be decreased in patients with diabetes as compared to those in normal controls. Our observations in the present study showed a definite lowering of serum chromium in diabetic patients (c=0.01), while in serum zinc level was found to be significantly lowered in diabetic (c=0.7) and in magnesium was (c=18). Hence, these trace element deficiencies appear to be an additional risk factor in the development and progress of disease and they contribute to the pathogenesis of diabetes mellitus and its complications. Their repletion may be an effective therapeutic intervention in prevention of the progression of the diabetes and its complications, along with a glycaemic control and control of other risk factors.

The concentration of zinc were slightly increased with ages especially the groups between (30-42) years and slightly reduced in the group between (49-60) years. The result observed that the concentration of chromium has the highest value in both sexes were in the age of (23-32) years. It was noted that the magnesium level was more in women than in men in the ages between (50-59) years.

The deficiency in zinc value in men and women is by 23.5% and 20.1% respectively. While the deficiency of chromium for men 40.1% and for women 40.4%. The mean Mg deficiency was 12.3%.

**Acknowledgement**

I would like to use the opportunity to great thanks first to the ministry of science and technology, especially material research office, medical tools centre and their staff for assistance in completing this study. It is with pleasure to me to express my special thanks and deep gratefulness to Dr. Fadhil M. Abid for his assistance.

**References**

Fig. (1) Phoenix-986 AA Flame Spectrophotometer

Fig. (2) Shimaadzu AA-6800 Flameless Atomic Absorption Spectrophotometer

Table (1): Instrument Conditions for Cr, Zn, and Mg Elements.

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Fig. (3): The Characteristic of Cr Element

Fig. (4): The Characteristic of Zn Element
Fig.(5): The Characteristic of Mg Element

Table (2) The Concentration of Cr, Zn, and Mg with their Absorbance's

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<th>Zn con. (ppm)</th>
<th>Mg con. (ppm)</th>
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