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Evaluation of status of toxic metals in biological samples of diabetes mellitus patients

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ABSTRACT

There is accumulating evidence that the metabolism of several trace elements is altered in diabetes mellitus and that these nutrients might have specific roles in the pathogenesis and progress of this disease. The aim of present study was to compare the level of toxic elements, lead (Pb), cadmium (Cd), and arsenic (As) in biological samples (whole blood, urine, and scalp hair) of patients having diabetes mellitus type-2 age ranged (31–60) ($n = 238$), with those of age matched non-diabetics (ND) as control subjects ($n = 196$), of both genders.

The concentrations of elements were measured by means of an atomic absorption spectrophotometer after microwave-assisted acid digestion. The validity and accuracy was checked by conventional wet acid digestion method and using certified reference materials. The overall recoveries of all elements were found in the range of 98.1–99.4% of certified values.

The results of this study showed that the mean values of Pb, Cd and, As were significantly higher in scalp hair samples of smoker and non-smoker diabetic patients as compared to control subjects ($p < 0.001$). The concentration of understudy toxic metals was also high in blood and urine samples of DM patient but difference was more significant in smoker DM patients. These results are consistent with those obtained in other studies, confirming that toxic metals may play a role in the development of diabetes mellitus.

1. Introduction

During the last few decades, endurance has consistently increased and lifestyle has become westernized in Pakistan, resulting in a dramatic increase in population with type-2 diabetes mellitus. Diabetes diagnoses are increasing at an alarming rate worldwide. The majority of diabetes-related deaths arise from vascular complications such as myocardial infarction, cerebrovascular disease, and peripheral vascular disease [1]. Khan and Ahmad [2] have reported 1.49% prevalence of diabetes in NWFP, Pakistan. In advance stages of diabetes, metabolism of protein and lipid is also altered.

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Many factors like heredity, age, obesity, diet, sex, sedentary life style, socio-economic status, hypertension and various stresses are involved in the etiology of DM.

Smoking has also been identified as a risk factor for insulin resistance, which can lead to diabetes. People with insulin resistance cannot properly use insulin and such people may initially have higher than normal amounts of insulin circulating in their blood, a condition known as hyperinsulinemia.

The clinical research suggests that the body's balance of trace elements and minerals can be disrupted by DM [3]. Conversely, research also suggests that early imbalances of specific elements may play an important role in upsetting healthy glucose and insulin metabolism [4]. Smoking delivers heavy metals (the term is used for some lighter metals and metalloids) to the lungs [5], particularly the more volatile metals such as cadmium and mercury that partition preferentially into the smoke phase on combustion [6]. Some of these readily pass into the bloodstream and may accumulate in specific organs [7]. Indeed smoking has long been considered a major source of several heavy metals in blood and various organs, and cadmium in particular is regarded as one of the "strong carcinogens" in tobacco smoke [8] with Ni, and As currently classified "carcinogenic to humans" by the International Agency for Research on Cancer (IARC) among 87 mainly organic carcinogens.

Diabetes mellitus is the most common cause of end-stage renal disease in many countries, accounting for about 40% of cases. The relationship between long-term environmental lead exposure and the progressive renal insufficiency in patients with type-2 diabetes and diabetic nephropathy is still unknown. Several epidemiological studies have demonstrated a positive association between blood lead levels and the age related decreases of renal function in the general population and suggested that environmental low-level lead exposure may accelerate the progression of renal function in the healthy people. In addition, environmental lead exposure may correlate to progressive renal insufficiency and lead chelation therapy or repeated lead chelation may improve and slow the progressive renal insufficiency in non-diabetic patients with chronic renal diseases [9].

From some cross-sectional population studies, it was reported that diabetes could augment the risk of cadmium-induced renal damage, especially tubular dysfunction [10]. It was consistence with previous study that diabetic patients may be more susceptible to the toxic effect of cadmium on the renal proximal tubule [11]. Several experimental studies have demonstrated an increased susceptibility toward cadmium nephrotoxicity [12] in spontaneously diabetic mice and hamsters, when compared with normal animals of the same strain. Streptozotocin-induced diabetic rats are more susceptible to cadmium nephrotoxicity than are normal rats when they are exposed subchronically to cadmium chloride in drinking water [12].

A dose–response relationship has also been observed between arsenic exposure and diabetes mellitus prevalence in community-based studies in high arsenic exposure areas in Taiwan [13] and Bangladesh [14], but not in several occupational studies on workers with high arsenic exposure. The features of diabetes mellitus observed in arsenic-exposed subjects in epidemiologic studies are actually similar to type-2 DM, which is characterized by a dual defect of both insulin resistance and a relative deficiency in insulin secretion [15].

Metal determination in human tissues is the most common application of biological monitoring for screening, diagnosis and assessment of metal exposures and their risks. Interest in the importance of trace elements to human health has increased considerably during last decades [16]. Among various biopsy materials, blood, scalp hair and urine may be used as bioindicators for this purpose. The low concentration of some trace elements in the aforementioned biological samples demands a technique with high sensitivity. Electrothermal (graphite furnace) atomic absorption spectrometry is usually the method of choice, and numerous reports testify to this observation [17].

This technique has need of solubilization of the analyte and complete or partial decomposition of the matrix using either convective systems or microwave ovens and dry ashing. The main advantage of microwave-assisted samples pre-treatment is its requirement of small amount of mineral acids and a reduction in the production of nitrous vapours. Microwave systems keep blank levels low because only small volumes of reagents are required and allow more samples to be processed per hour than conventional digestion systems [18].

As the rate of mortality is also increased in Pakistan during the last few decades due to diabetes mellitus, even though an extensive list of risk factors has been well characterized in its pathogenesis, tobacco smoke is considered the most important cause of diabetes mellitus, through active or passive smoking. Very limited data are available in Pakistan to show the association of diabetes mellitus with carcinogenic metals. This follow-up study of three years is aimed to evaluate the concentration of Cd, As and Pb in whole blood, urine and scalp hair among male smoker diabetic patients and healthy controls or referents (smoker and non-smoker) of same age group, from Hyderabad, Sindh, Pakistan. The biological samples were digested by microwave prior to analysis by atomic absorption spectrometer.

2. Material and methods

2.1. *Apparatus*

A PerkinElmer model A. Analyst 700 (Norwalk, CT) atomic absorption spectrometer equipped with deuterium background correction. The hollow cathode lamps of As, Pb and Cd were run under the conditions suggested by the manufacturer (Table 1). Integrated absorbance signals computed by the AA spectrometer were employed throughout. A Pel (PMO23) domestic microwave oven (maximum heating power of 900 W) was used for digestion of the biological samples. Acid-washed polytetrafluoroethylene (PTFE) vessels and flasks were used for preparing and storing solutions.

2.2. *Reagents and glass wares*

Ultrapure water obtained form ELGA labwater system (Bucks, UK), was used throughout the work. Concentrated nitric acid 65% and 30% hydrogen peroxide purchased from Merck