# Analytical Procedure for Determination of Lead in Blood and Urine by Atomic Absorption Spectrometry

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## ABSTRACT

Lead is one of the natural constituents of earth found in soils, plants, and water. Lead predominantly a concern for neurological toxic effect particularly in children due to its irreversible neurological damage. The common spectroscopic methods in trace element analysis in biological fluids are dithizone extraction, polarography, spectral analysis, atomic absorption spectrometry (AAS), inductively coupled plasma atomic emission spectrometry. A graphite furnace atomic absorption spectrophotometer is a valuable, simpler, and cost-effective spectrometric technique for the identification of lead absorbance in the human system. The GF-AAS method can be employed for the measurement of lead percentage in whole blood and urine, and the results acquired are helpful in biological monitoring and clinical diagnostic of work related to surrounding lead exposure. The paper reports the average lead concentration, and calibration graphs for standards as well as absorbance against lead concentration, which will be productive in future for analytical determination of lead in blood and urine.

Keywords: analytical method, atomic absorption spectroscopy, blood, graphite furnace, lead, trace elements, urine.

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### I. INTRODUCTION

Human organisms are considered as complex multielement dynamic systems consisting of about 65 to 80 elements. These elements can be classified based on two factors: 1) On chemical analysis, and 2) Based on their role in the human body system. Depending on chemical analysis, these elements can be subdivided into three groups namely: macro-elements, trace-elements, and ultra-trace elements; while depending on the element role in the biological system, they are divided into four types namely: organic, inorganic, potentially toxic and toxic trace elements [1]. Blood is an essential constituent of a human organism, which mainly consists of two components, plasma, and blood corpuscles. Blood corpuscles are erythrocytes, thrombocytes, and leukocytes. Plasma is highly made up of water and is a solution of dissolved organic and inorganic compounds in water [2]. Organic proteins are albumin, globulins, and fibrinogen, while inorganic components are salts of sodium. In addition, vitamins, nutrients, inorganic ions, enzymes occur in plasma. Moreover, human urine is an inhomogeneous medium consisting of nitrogen products such as urea, creatinine, amino acids, protein and glucose and also salts of sodium, calcium ammonium and magnesium [3]. Analysis of blood and/or urine is pronounced and the analysis of trace element percentage in blood and urine is beneficial in clinical and biological studies. Researchers prefer analysis of blood, rather than urine, because urine is an excretion material and contains a higher amount of salt and its derivatives. High content of salt and its derivatives in urine tends to produce undesired, non-repeatable and non-reliable analytical outcome and is considered to be more problematic when compared to the analysis of blood [4]. In general, biological fluids are considered as complex matrix composition susceptible to contamination, while handling, sampling, and conservation. For instance, a slight hemolysis may result in an increase in the concentration of iron, copper, zinc, lead and manganese in blood serum. Furthermore, medical syringes and anticoagulants may contaminate the sample at the stage of conservation [5].

The presence of element traces in biological fluids is necessary for its proper and desired function, while the presence of some non-essential elements can pose a risk of toxicity. Both essential and non-essential elements can be toxic, if concentrations exceed a certain threshold [6].

Lead is one of the natural constituents of earth found in soils, plants and water. As a result of its mining and its use in synthesis of daily products caused pollution in the environment, making it highly toxic. Lead acts as a catalyst in the process of oxidative tissue damage, due to its strong binding capacity and interferences with enzymes and structural proteins, which is evident by many studies describing biomarkers of lead toxicity. However, few concerns with the exposure to lead are decrease in erythrocyte, basophilic, increase in urinary acid. The most common signs of toxicity of lead are evident among all age group with wide variety of symptoms such as high blood pressure, muscle and joint pain, miscarriage, premature birth, lower birth weight, retarded growth and brain development, and learning difficulties [7].

Lead is predominantly a concern for neurological toxic effects particularly in children; in some cases, it is too serious such as irreversible neurological damage. The difficulty in diagnosis of lead toxic is due to the asymptotic symptoms of the affected individual is relatively uncertain, while the only trusted way to diagnose the exposure of lead by an individual is with whole blood lead analysis. In addition to blood and urine, one may analyze teeth, bone and hair as the most utile tool for screening and diagnosis of lead toxicity [8].

# II. THEORETICAL ASPECT AND INSTRUMENTATION

are numerous analytical methods for the identification of trace elements in samples such as X-ray techniques, Potentiometry, Atomic spectroscopy, Voltammetry and Nuclear techniques. A comparison of these methods is presented elsewhere [9], which will help in selecting the suitable method depending on the specification of the sample or the element which is to be analyzed. The experimental arrangement of GF-AAS and its theoretical aspect such as determination of concentration, examining the molecule property using Beer-Lambert law is mentioned in detail elsewhere [10].

#### III. MATERIALS AND METHODS

The selection of a spectroscopic method for element analysis is based on the process of sample preparation and sensitivity of the material to the exciting energy. Most common spectroscopic methods in element analysis of biological fluids are dithizone extraction, polarography, spectral analysis, atomic absorption spectrometry (AAS), inductively coupled plasma atomic emission spectrometry (ICP-AES) and inductively coupled mass spectrometry (IC-MS) [9], [10].

Trace elemental analysis with a graphite furnace atomic absorption spectrophotometer (GF-AAS) is mentioned [9]-[11]. For blood analysis, a sample of 200 µl is diluted in 1.6M solution of 65% of HNO<sub>3</sub> then centrifuged for about 7 to 10 minutes at 3000 rpm. Furthermore, for urine analysis, a sample of 9 ml is diluted 1 ml solution of 65% of HNO<sub>3</sub> then centrifuged for about 15 to 20 minutes at 3000 rpm. The supernatant of analyte is filtered and then can be used in GF-AAS system at wavelength 283.3 nm, slit of 0.5 and lamp current of 10 mA. Different standard solutions  $10 \mu g/ml$ ,  $20 \mu g/ml$ ,  $50 \mu g/ml$  and  $100 \mu g/ml$ may employed at atomization temperature up to 2000 °C. The average concentration of lead in the analyte is as shown in table I, while Fig. 1 shows the calibration curve of standard solutions with averaged lead content, and. Fig. 2 shows the linearity between the absorbance and the lead concentration.

TABLE I: AVERAGE LEAD CONCENTRATION FOR THE STANDARD SOLUTIONS CONCENTRATION

BOECHOILD CONCENTRATION		
	Standard Solutions	Average Lead
	Concentration	Concentration
	$(\mu g/ml)$	(μg/ml)
	10	$10.54 \pm 1.06$
	20	$21.70 \pm 0.53$
	50	$52.14 \pm 2.81$
	100	$100.09 \pm 1.40$

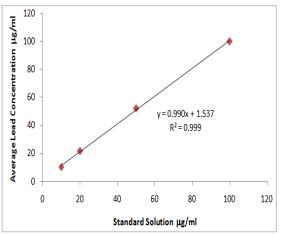


Fig. 1. Calibration curves of standard solutions and average lead concentration.

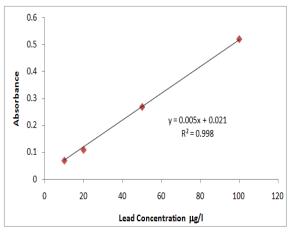


Fig. 2. Absorbance vs. Lead concentration.

### IV. DISCUSSION AND CONCLUSIONS

The toxic effect of lead exposure in humans has been a concern in the past few decades. A small amount of lead in the human body is vulnerable to affect physical as well as mental health. Toxicity of lead occurs when lead builds up in the human body due to lead-contaminated dust in aged buildings or due to lead-based paint or contaminated air, water or soil. Lead poisoning affects multiple body systems such as cardiovascular, neurological, gastrointestinal, and hematological systems. Chronic lead exposure can cause headache, muscle weakness, allergy, convulsions and paralysis, while acute lead exposure may cause nausea, vomiting and abdominal pain sometimes leading to convulsion and sometime leading to death.

Laboratory diagnostic and screening studies of trace elements in biological fluids can be simple and more effective and can help diagnose metal toxicity and nutritional deficiencies. The preferred methods for such studies are AAS with the Zeeman correction. Whole blood and urine lead level are the most widely used method to determine the absorbed dose in the human system. GF-AAS is simpler, quicker, repeatable, reliable, and cost-effective spectrometric technique for the identification of lead absorbance in human system. The GF-AAS is a valuable method for biological monitoring and clinical diagnostic of work related or surrounding lead exposure. The reported average lead concentration, and calibration graphs for standards as well as absorbance may be useful for analytical determination of lead in blood and urine.

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