**Salahaddin Univeristy**

**College of Science**

**Environmental Science and Health Department**

**Comparative Heavy Metals (Ni, Cd and Pb) Blood Between Smokers and Non-Smokers**

**Research project**

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**ABSTRACT**

**Keywords:** **Heavy metal, Blood, Smokers, Non-Smokers.**

**1.INTRODUCTION**

Tobacco is a major contributor to some metal intake in humans. Researchers studied the concentrations of Pb, Cd, Ni and other metals in the blood of an urban population. It was reported that tobacco smoking is a common source of Pb and Cd exposure. (Oehme & Lund. 1979).

The consumption of tobacco products and number of smokers have been increasing steadily all over the world. The use of cigarettes constitutes one of the major causes of morbidity and mortality in the world. In the tobacco plantation herbicides, insecticides, and fungicides are used to control the various parasites and plant diseases. Tobacco smoke has toxic, genotoxic, and carcinogenic properties. The cigarette smoke contains both organic and inorganic human carcinogenic compounds. Containing 4000 identified chemical compounds, cigarette smoke is very harmful and toxic for human health. Of these toxic materials are heavy metals, particularly cadmium and lead inhaled through.

Several heavy metals found in tobacco smoke such as Cd, Cr, Pb, and Ni also accumulate in tissues and fluids through smoking Tobacco smoking is the most important single source of Cd exposure in the general population. According to the most important sources of Cd in humans are smoking and food. Cadmium can enter the body through tobacco smoking, diet, drinking water, and inhaling it from the air. Small amounts of Cd taken over many years may cause kidney damage and fragile bones, since Cd is mainly stored in bone, liver, and kidneys.

Furthermore, Cd causes stomach irritation, vomiting, and diarrhea. Cadmium and lead, present in tobacco smoke, contribute substantially to cancer risk . (Pappas, R. S. et al. 2007)

Cadmium is a group I carcinogen and lead has recently been elevated from a group IIB to a group IIA carcinogen . Cigarette smoke contains substantial amounts of Cd. Average Cd levels in cigarettes range from 1000 to 3000 *μ*g/kg. One pack of cigarettes deposits 2–4 *μ*g into the lungs of a smoker while some of the smoke passes into the air to be inhaled by smokers and nonsmokers alike , which means, for 20 cigarettes smoked, approximately 2–4 *μ*g of Cd is inhaled by the smoker and as much as a microgram of Cd spreads into the environment. Mussalo-Rauhamaa et al. reported that the mean contents in filter cigarette tobacco sampled from Finland were 1.7 and 2.4 *μ*g*−*1 (dry weight) for Cd and Pb, respectively.

Lead is a highly toxic metal and is capable of causing serious effects on the brain, nervous system, and red blood cells. An increase of Pb level is associated with a decrease in the intelligence quotient (IQ) levels and potential behavioral problems. A survey of middle-aged men in 24 British towns showed a strong association between blood Pb concentrations and alcohol and cigarette smoking. Smoking of 20 cigarettes a day has been estimated to result in the inhalation of 1–5 *μ*g Pb . The WHO estimates 2–6% of Pb in cigarettes is inhaled by the smoker . It was reported that Pb in tobacco has been associated with impaired fetal growth and brain development.( Sani, A. et al. 2017)

Nickel (Ni) is the 24 th most abundant element in Earth’s crust. On the one hand, at a low concentration, Ni is a nutritionally essential trace element for animals. Ni is also a required trace element for several enzymes that play critical roles in energy and nitrogen metabolism. On the other hand, at a high concentration, Ni is a toxic element . The unique physical and chemical properties of Ni make it and its compounds suitable materials for many applications widely found in modern industries. The widespread use of Ni increases its concentration in biogeochemical cycles and enhances human exposure to it and its compounds through environmental contamination and occupational exposure. Human exposure to Ni occurs primarily via inhalation, ingestion, and dermal absorption. In humans, Ni has been known to cause liver, kidney, spleen, brain, and tissue damage. The International Agency for Research on Cancer (IARC) has classified Ni compounds such as nickel sulfate (NiSO4), nickel oxide (NiO), nickel hydroxides, and crystalline nickel as carcinogenic agents to humans.( Vacchina et al.2014)

In Northern Ireland, around 340,000 people aged 16 and over smoke. Smoking contributes to not only many cancers, heart disease, bronchitis and asthma, but other illnesses, including stroke, which causes around 2,400 deaths per year here, all of them avoidable.( [Hongrui Guo](https://pubmed.ncbi.nlm.nih.gov/?term=Guo%20H%5BAuthor%5D) et al.2019)

The consumption of tobacco products and the number of smokers have been increasing steadily throughout the world and Saudi Arabia is no exception to this. Tobacco in Saudi Arabia continued to display high growth in 2010, rising significantly in both volume and value terms. (Kristiansen et al. 1997).

The term heavy metal refer to those metals that are having toxic effects on human physiology and their exposure can result in significant abnormality of human health. There are varieties of metals that can be considered as heavy metals such as lead ,mercury ,cadmium which are never desirable for human physiology but some other metals like zinc , iron have some benefits to maintain good human health. On other hand limited intake of copper ,manganese , chromium ,and molybdenum are also important in the human diet. The toxicity of heavy metals depend on a number of factors like total amount absorbed and whether the exposure of heavy metals was acute or chronic for example ,the elemental mercury is relatively target the gastrointestinal tract and also poorly absorbed through skin. Inhalation or injection of element mercury may also have some disastrous effects. (Syed fahad et al.2013).

In the one of researches in saudi Arabia , he talks about smoking and the source of heavy metals in this way , The toxic or heavy metals enter the human body through many sources, namely, the houses paint, fish, dental amalgam, farming, mining, and smoking including second hand smoke.

Smoking is an important source of exposure to nickel, cadmium , lead, and other toxic metals. The cigarette smoke contains such toxic heavy metals inhaled. Once inhaled through smoking, toxic metals have a long biological life span. Chronic adverse effects on human health may, therefore, in later years result from prolonged intake of such toxic elements, some of which are powerful carcinogens. Based on their extreme toxicity, toxic and heavy metals must be detected at very low levels in the human fluids and tissues .In this context, blood and hair are the most suitable human biological samples generally used in such metal analysis. The blood samples have been used to assess the heavy metals levels in human body for several years. Due to the partitioning of the toxic and heavy metals in the hair, recently hair samples have been used as good index in such studies. (Majid alrobaian et al. 2019).

The people who are smoking too much they confront:shortness of breath, coughing, fatigue, sleep and heart problems, weight loss, and depression.

The aim of the study was to assess the heavy metal concentrations in smoker and nonsmoker blood samples.

The objectives of the present study were:

1. To investigate heavy metal concentrations in bloods of smokers and nonsmokers.

2. To find out if there are significant differences between blood heavymetal contents of smokers and nonsmokers.

**2. MATERIALS AND METHODS**

**2.1. Sampling and method Description**

**Conventional Wet Acid Method** was used for blood sample digestion. Whole blood was taken into small beakers for testing (Ni, Pb and Cd) (Yahaya et al., 2013).

The sample is the blood that taken from 6 person, 4 are smokers and 2 of them are non-smokers (controls), also with 2 blank samples.

**2.2. Sample Collection and Storage**

First, we taken 4 blood samples from 4 people that are smokers, they are smoking for long time, Also we taken 2 blood samples from 2 people that are non-smokers.

Also we prepare 2 blank samples that contain Nitrate acid and hydrogen peroxide.

After collection samples we storage in the EDTA tubes at the 4Cᵒ.

**2.3. Heavy Metals Extraction and Determination**

**Conventional Wet Acid Method** was used for blood sample digestion (Yahaya et al., 2013).

***Pretreatment of sample containers:***All glassware and sample containers (polyethylene plastic bottles) and Teflon beakers were thoroughly washed with non-ionic detergent solution to ensure all dirts were removed followed by rinsing with tap water until free of detergent.

These were thoroughly rinsed with distilled water followed by soaking in 10% (v/v) nitric acid for 48 hours and finally rinsed with triply distilled water and dried in the oven at 800 C prior to sampling and analysis.

***Digestion using Conventional Wet Acid Method (CDM):***Blood samples were digested by the Conventional Wet Acid Method by adopting the method of Memon et al. (2007). Accurately 0.5 mL of whole blood was taking into Pyrex flask separately. To this was added 3 mL of freshly prepared mixture of concentrated nitric acid and hydrogen peroxide [HNO3 - H2O2] (2:1V/V) and stood for 10 minutes. The flasks were covered with watch glass and then digested at 60 - 70 0C for 1 – 2 hours. The digests were then treated with 2 mL nitric acid and few drops of H2O2, while heating continued hot plate at about 80 0C until a clear digested solution was obtained. The excess acid mixture was evaporated to semi - dry mass, cooled and diluted with 0.1 mL nitric acid. These were transferred into 100 mL volumetric flask and diluted to mark using triply distilled water. A blank extraction (without the sample) was carried out through the complete procedure using triply distilled water. The concentrations of heavy metals (Cd, Ni, and Pb) were determined with an Atomic Absorption Spectrophotometer (AAS). Standard solutions of each metal (Cd, Ni, and Pb) were prepared according to the procedure for AAS to be used for the calibration curve (Shaltout et al., 2007).

**2.4. Digestion of whole blood**

65%nitric acid was added to the blood sample (3ml) that was stored previously at 4◦Cfor analysis. incubator was used for the digestion of the blood samples. For digestion of the samples, 1 ml of the sample and 3 ml of a mixture of concentrated hydrochloric acid and nitric acids in (1 ml 30% hydrogen pyroxide H2O2 , 1 ml 65% Nitric acid HNO3) were transferred into a 125 ml pyrex conical flask. The samples were digested for 1-2 hours. The digestion was stopped when a colourless solution was obtained and then it was evaporated to dryness. The solution was diluted to 100 ml with distilled water.

**4. RESULT**

**5. DISCUSSION**

**6. CONCLUSION**

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