Biological Monitoring Heavy Metals in Fingernails and Scalp Hair of Autoworkers in Saudi Arabia

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Abstract

Biological materials have gained an irreplaceable position as a bioindicator for environmental pollution with heavy metals. Human hair and nails have come to play an important role in this subject according to the recommendations by the World Health Organization (WHO). The levels of selected toxic and heavy metals (Pb, Zn, Cd, and Ni) in the fingernails and scalp hair of autoworkers in Saudi Arabia were estimated to study exposure to chemical pollutants metals. The samples were collected from different ages of male autoworkers (car mechanics, auto welding, repair car tires washing, and oil change services center) and office workers (control) volunteers in Hail city. The analysis was performed using inductively coupled plasma mass spectrometry (ICP-MS) after microwave acid digestion at their recommended conditions. The data obtained from office workers (control) samples were used for this purpose. A systematic analysis investigation was done for all samples taking into consideration the age and experience per year. As a result of this research, it has been revealed that there was a statistically significant relationship between the type of work (autoworkers) or workplace and the heavy metal concentration detected in fingernails and scalp hair samples, which represent a very important guide for the labor and health organizations. This result proved that fingernails and scalp hair could be used as a good biological indicator for the assessment of toxic heavy metal pollution.

Keywords: Fingernails, Scalp hair, Autoworkers, Saudi Arabia, Heavy metal pollution

Introduction

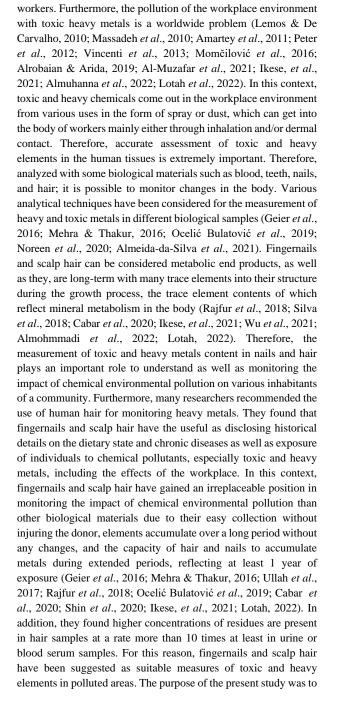
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Environmental pollution has an important role in increasing the levels of toxic and heavy metals in the human body, which lead to various chronic diseases (Rajfur *et al.*, 2018; Silva *et al.*, 2018; Ocelić Bulatović *et al.*, 2019; Cabar *et al.*, 2020; Noreen *et al.*, 2020; Almeida-da-Silva *et al.*, 2021; Ikese, *et al.*, 2021; Wu *et al.*, 2021; Lotah, 2022). The working environment has come to play an important role in this subject, due to the direct relationship between man and the degree of hazards chemical agents present in their workplace environment. In addition, each workplace environment

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is not unique but also in the degree of hazards it poses to the

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find out baseline information about the level of toxic and heavy elements (Pb, Zn, Cd, and Ni) in fingernails and scalp hair of autoworkers in Saudi Arabia using ICP-MS. No information has been reported and/ or available to evaluate the exposure of these metals in autoworkers in our area. Within this context, our study mainly aimed at the relationship between the workplace, such as autoworkers, and the presence of toxic and heavy metal levels in the hair and nail. Therefore, this study focused on the effect of autoworker's workplace and years of working on the accumulation of toxic and heavy metals in the hair and nails of workers. On the other hand, the auto workers chosen for this study were in direct contact with thinner, grease, petrol, diesel, etc. The obtained data can be lead to develop and help to find an easy and better diagnostic assessment for heavy metals toxicity particularly as a screening program for autoworkers and others.

Materials and Methods

All reagents used in this work are AR grade and there is no need for further purification. Deionized water with 18 M Ω cm-1 was used to prepare all standard solutions, dilute samples, and wash all tools and glassware throughout. Hydrogen peroxide (H₂O₂, 35%), and nitric acid (HNO₃, 69%) were obtained from Sigma-Aldrich. All plastic and glassware used were washed and cleaned with dilute nitric acid, and air-dried before use.

Samples Collection

Samples in this study were collected from different sites in Hail city, the northern province of Saudi Arabia. The fresh samples (fingernails and scalp hair) were collected from 83 exposed subjects (autoworkers) and 24 office workers (control) using sterilized scissors and nail clippers of volunteers across several workplaces at the age of 18 to 60 years old males. All the individuals are not allowed to cut their fingernails and scalp hair for about 60 days before sampling. The study was conducted in accordance with the Declaration of Helsinki and approved by the Research Ethics Committee of Hail University, Hail, Saudi Arabia. Scalp hair samples were obtained from workers who did not have colored or treated hair and taken from the nape of the head as near as possible to the scalp. Nail samples were collected from all 10 fingers. In this context, all samples were cut into pieces as small as possible. Samples were classified according to the type of workplace, stored at about 5°C until digestion and analysis processes. In this context, samples were collected from healthy volunteers who unpolluted workplace sites (control), for purpose of comparison. Before samples collection, the hands of all autoworkers were craftily washed with medicated soap metal-free and then dried to remove any external contamination by using sterile tissue paper. In order not to affect the determination data, care was taken not to have the hair dyed while collecting the samples for study. The collected samples were kept to seal tightly coded polythene bags in our laboratory for pre-treatment. Before sample collection, questionnaire surveys were performed for all participants containing highlights of information such as age, home address, nutritional habits, smoking water source, total monthly income, current health with regard to chronic diseases or consequently drugs treatment, smoking habits, and occupational

exposure to heavy metals. Information was obtained through multiple-choice questions.

Washing Procedure

The key aim of the washing process is to remove the external pollutants from hair and nails pre-digestion technique to have a true level of endogenous metals. In the other words, it will eliminate only the surface external pollutants without extracting elements from the samples or depositing elements on them. The results of existing and previous research have indicated that detergent washing is not suitable, where it reduced the metal concentrations of samples compared with the organic solvent washing. However, washing procedures do not essentially affect the heavy metals concentrations due to the strong complex of disulfide groups in the keratin proteins with these metals (Pozebon et al., 2017; Alrobaian & Arida, 2019; Abulhamael et al., 2022). Fingernails and scalp hair samples were initially washed with noionic detergent, Triton X-100, and then by CH₃COCH₃ (acetone), after which washed by deionized H2O. After washing processes, the fingernails and scalp hair samples were dried in an electric oven at about 60°C and protected in polyethylene bags until analysis. According to the International Atomic Energy Agency (Gönener et al., 2020; Lotah et al., 2022; Suragan & Durmuşkaya, 2022), this method has been come to be one of the most successful washing procedures in removing external trace elements and preserving internal trace elements.

Sample Preparation for Trace Element Analysis

After the drying process, each investigated sample (0.5 g of fingernails or scalp hair) placed in a Teflon container was individually digested in the microwave digestion system with a mixture of 7 mL of concentrated HNO3 and 3 mL of H2O2. In this system, the temperature ramped to 200°C for 10 minutes, after which held for10 minutes, and then cooled to reach room temperature. This method of digestion includes minimum sample handling steps which lead to minimizing the sample contamination besides a high large number of samples, which lead to improving the overall throughput and the reliability of the suggested analysis protocol. The microwave procedure operated according to the report by the US Environmental Protection Organization (USEPA) (Alrobaian & Arida, 2019). After cooling of the digestion vessel, the resulting clear solution was quantitatively diluted with deionized water and an aliquot was inserted into the cell for analysis using ICP-MS. A blank sample, containing deionized water was also used under the same conditions. The investigated toxic and heavy metals were measured by inductively coupled Plasma mass spectrometer- ICPMS2030 (Shimadzu- Japan) under optimized plasma conditions in all digested samples and blanks as well. The concentration of the investigated metals was measured automatically using the standard graph, which was prepared under the same plasma conditions. The system was adjusted to triplicate determined of the samples as well as the automatically calculated both correlation coefficient and the relative standard deviation, which found to be 0.99998 and < 2 %, respectively. The measurement method (ICP-MS) was validated using working calibration solutions of the under-investigated metal ions (Pb, Zn, Cd, and Ni). Standard solutions of all the metals investigated were prepared using stepwise dilutions of certified standard stock solutions. These solutions for calibration of all the metals investigated were prepared from ultra-grade a 1000 mg/L ICP multielement solution (Merck, Milan, Italy), using stepwise dilutions of certified standard stock solutions. The procedure validation included linearity, accuracy, and precision.

Results and Discussion

Each work environment is unique and the degree of hazards it poses to the workers. This study revealed the risks of toxic and heavy metals coming from highly polluted environments and the workers in these places are directly in contact with various pollutants and have different health problems. Heavy metal pollution in the workplace environment has come to be a worldwide problem. In this context, the use of nails and hair analysis for toxic and heavy metals has been recommended by the World Health Organization (WHO), due to the dependence of toxic and heavy metals levels in them on environmental exposure. In this work, we determine the concentration of toxic and heavy metals (Pb, Zn, Cd, and Ni) in fingernails and scalp hair samples of volunteer workers and controls (no direct metal exposure) by ICP-MS technique to estimate any relation between heavy metals concentration and environmental and/or occupational exposure. The ICP-MS measurement was validated by analysis of various standard samples and spiking are known concentrations of toxic and heavy metals, which showed good accuracy, linearity, precision, and lower limit of detection. The measurements were performed in triplicate and the mean was automatically calculated. Many researchers reported that there is a relationship between the concentration of trace elements in human tissues and many dangerous human diseases such as cancer, mutagenicity, neurobehavioral decrements, cardiovascular diseases, gastrointestinal irritation, and immune dysfunction which maybe lead to deaths (Pozebon et al., 2017; Gönener et al., 2020; Sumantri et al., 2022).

Nails and Hair Samples Analysis Results

In the current study, bio-monitoring of four heavy metals was assessed by ICP-MS after microwave-assisted sample digestion with HNO₃ in the whole nails and hair samples (total n=107) volunteer workers and controls (no direct metal exposure). The volunteers were divided into two groups: control (n=24, 22.4 %), and workers (n=83, 77.6 %). In this context, the volunteer workers were categorized according to the types of occupations into four sets, namely, car mechanics workers (n= 21), and auto welding workers (n=20), repair car tires workers (n= 22), and washing and oil change services center workers (n=20). For comparison, the tested metals (Pb, Cd, Zn, and Ni) were also measured in whole fingernails and scalp hair samples (n=24) of control. The control volunteers were classified according to the ages as well as exposure duration, Year (y) as shown in Table 1. The concentration ranges and averages of Cd, Ni, Zn, and Pb in fingernails of unpolluted workplace site (control) are 0.28 -0.56(0.38), 4.67 -10.12(7.04), 57.34 - 107.44 (81.87), and 8.45 -12061(10.27), while for scalp hair samples are 0.10 - 0.43 (0.23), 3.12 - 9.14(6.01), 50.56 - 105.61(73.84), and 6.54 - 13.90(9.41), respectively, Table 1. These obtained results are consistent with that reported for other parallel studies and are they are agreement the international tolerance levels (Nakaona et al., 2020; Yin et al., 2021). In this context, the levels of these toxic heavy metals in a biological specimen such as fingernails and scalp hair should be below the international tolerance levels as far as possible. However, environmental Pollutants have come to play an important role to introduce these elements into human biological systems, and therefore the term tolerant levels should be used instead of reference values (Alrobaian & Arida, 2019; Nakaona et al., 2020). On the other hand, the obtained results show that the levels of the studied metals of fingernails and scalp hair in unpolluted workplace site volunteers reasonably fluctuated within a relatively narrow range for a given element. Indeed, the relative fluctuation in the obtained values in each studied metal level reflects the variety in culture, age, environment, and habits (Nakaona et al., 2020).

Table 1. Heavy metals levels (μ g/g) in fingernails and scalp hair samples of unpolluted workplace site (control)

| No | Exposure duration, | A aa* | C | Cd | Ν | Ji | Z | 'n | Pb | | |
|----|--------------------|------------------|-------|-------|-------|-------|--------|-------|-------|-------|--|
| No | Year(y) | Age [*] | Nails | Hairs | Nails | Hairs | Nails | Hairs | Nails | Hairs | |
| 1 | 1 | а | 0.39 | 0.18 | 5.23 | 4.76 | 61.47 | 72.31 | 8.45 | 6.54 | |
| 2 | 2 | а | 0.32 | 0.20 | 4.87 | 3.12 | 57.34 | 50.56 | 9.33 | 11.30 | |
| 3 | 4 | b | 0.28 | 0.19 | 7.24 | 6.68 | 78.35 | 69.57 | 8.97 | 6.86 | |
| 4 | 4 | b | 0.29 | 0.19 | 4.67 | 3.42 | 94.24 | 80.57 | 11.43 | 8.59 | |
| 5 | 5 | b | 0.31 | 0.24 | 6.45 | 4.96 | 63.26 | 47.83 | 9.47 | 11.45 | |
| 6 | 7 | b | 0.35 | 0.17 | 6.86 | 5.39 | 78.35 | 80.56 | 9.45 | 11.98 | |
| 7 | 9 | b | 0.40 | 0.13 | 8.32 | 8.21 | 81.34 | 69.58 | 10.34 | 8.45 | |
| 8 | 9 | b | 0.32 | 0.11 | 6.78 | 7.12 | 90.85 | 83.35 | 9.46 | 7.67 | |
| 9 | 10 | b | 0.28 | 0.19 | 5.91 | 4.35 | 84.74 | 68.34 | 8.85 | 10.79 | |
| 10 | 10 | с | 0.42 | 0.31 | 6.47 | 3.67 | 90.07 | 78.48 | 12.23 | 7.56 | |
| 11 | 10 | с | 0.39 | 0.25 | 5.86 | 6.33 | 107.44 | 90.35 | 10.36 | 8.98 | |
| 12 | 10 | с | 0.48 | 0.32 | 8.33 | 5.95 | 88.25 | 93.47 | 8.58 | 5.97 | |
| 13 | 12 | с | 0.29 | 0.21 | 7.45 | 5.92 | 78.91 | 56.79 | 9.85 | 7.93 | |
| 14 | 12 | с | 0.28 | 0.10 | 4.79 | 4.23 | 68.36 | 54.88 | 10.47 | 8.85 | |
| 15 | 15 | с | 0.44 | 0.22 | 7.60 | 6.87 | 70.34 | 78.63 | 8.49 | 10.76 | |

| 24 | 38 | e | 0.51 | 0.36 | 10.12 | 8.93 | 87.98 | 99.44 | 12.97 | 9.93 |
|----|----|---|------|------|-------|------|-------|--------|-------|-------|
| 23 | 37 | e | 0.49 | 0.31 | 9.94 | 7.69 | 92.36 | 105.61 | 10.31 | 13.90 |
| 22 | 30 | e | 0.56 | 0.41 | 8.58 | 9.14 | 98.56 | 75.30 | 12.54 | 8.56 |
| 21 | 27 | d | 0.34 | 0.19 | 6.15 | 5.98 | 90.35 | 76.84 | 9.37 | 10.89 |
| 20 | 22 | d | 0.28 | 0.11 | 5.49 | 4.87 | 79.48 | 64.84 | 11.62 | 9.97 |
| 19 | 20 | d | 0.46 | 0.35 | 8.95 | 7.23 | 69.37 | 54.61 | 9.83 | 7.85 |
| 18 | 18 | d | 0.42 | 0.23 | 8.98 | 9.12 | 87.34 | 60.86 | 11.84 | 8.77 |
| 17 | 16 | c | 0.31 | 0.15 | 7.55 | 5.49 | 75.85 | 80.98 | 10.38 | 12.87 |
| 16 | 15 | с | 0.51 | 0.43 | 6.44 | 4.86 | 90.36 | 78.57 | 11.86 | 9.34 |

* Age: $a = \le 20$ y, b = 21-30 y, c = 31-40 y, d = 41-50 y, e = 51-60 y.

Table 2 shows the concentration of toxic and heavy metal (Pb, Cd, Zn, and Ni) in fingernails and scalp hair samples of 21 car mechanics workers volunteers. It was found that the concentration ranges and averages of Cd, Ni, Zn, and Pb in fingernails of car mechanics workers were 0.41-0.80 (0.63), 4.12-11.32 (7.96), 62.8-142.21 (95.66), 6.75-19.43(13.29), while the concentration ranges and averages of Cd, Ni, Zn and Pb in scalp hair of car mechanics workers were 0.12 - 0.76 (0.38), 4.10-13.20 (9.73), 72.65-142.21 (112.43), and 7.12-25.32(17.93), respectively.

From **Table 3**, it was found that the concentration ranges and averages of Cd, Ni, Zn, and Pb in fingernails of car mechanics workers were 0.35- 0.72 (0.49), 7.94 - 14.96 (11.07), 83.65 - 150.70 (109.78), and 7.10 - 17.54(12.27), while the concentration ranges and averages of Cd, Ni, Zn and Pb in scalp hair of car mechanics workers were 0.39 - 0.65 (0.50), 8.21-13.27 (10.90), 88.71-16121 (126.95), and 7.82 - 19.21(14.27), respectively.

The investigated toxic heavy elements were also determined fingernails and scalp hair samples of 22 repair car tire workers volunteers. The data obtained are summarized in **Table 4**. It was found that the concentration ranges and averages of Cd, Ni, Zn, and Pb in fingernails of car mechanics workers were 0.38-0.80 (0.61), 9.20-15.96 (12.88), 61.5-119.61 (97.78), and 6.98-16.86(11.65), while the concentration ranges and averages of Cd, Ni, Zn and Pb in scalp hair of car mechanics workers were 0.41-0.91 (0.73), 8.79-16.88 (12.99), 66.21-126.51 (101.09), and 7.54-15.94(12.03), respectively.

Table 5 summarizes the concentration of the investigated toxic and heavy metals fingernails and scalp hair samples of 20 washing and oil change services center workers volunteers. The concentration ranges and averages of Cd, Ni, Zn and Pb in fingernails of car mechanics workers were 0.43 - 0.71 (0.55), 5.43 - 17.95 (11.81), 72.6 - 144.6 (104.37), and 7.98 - 18.76(12.07), while the concentration of theses metals hair samples were 0.48 - 0.87 (0.66), 6.32 - 18.79 (12.49), 77.43 - 133.62 (108.58), and 8.32 - 20.21(14.68), respectively.

Table 2. Heavy metals levels $(\mu g/g)$ in fingernails and scalp hair samples of car mechanics workers

| No | Exposure duration, | 1 22 | (| Cd | Ν | Ni | Zn | | Pb | |
|----|--------------------|------|-------|-------|-------|-------|--------|--------|-------|-------|
| No | Year(y) | Age | Nails | Hairs | Nails | Hairs | Nails | Hairs | Nails | Hairs |
| 1 | 2 | а | 0.41 | 0.21 | 5.81 | 4.77 | 62.8 | 81.30 | 8.21 | 7.12 |
| 2 | 3 | b | 0.44 | 0.12 | 4.88 | 4.10 | 71.24 | 79.73 | 6.75 | 7.79 |
| 3 | 3 | b | 0.41 | 0.14 | 5.76 | 5.13 | 78.94 | 72.65 | 7.42 | 10.34 |
| 4 | 4 | b | 0.53 | 0.16 | 4.12 | 6.75 | 70.57 | 86.77 | 7.89 | 8.98 |
| 5 | 5 | b | 0.57 | 0.14 | 5,72 | 6.81 | 90.23 | 90.55 | 9.31 | 13.43 |
| 6 | 6 | b | 0.47 | 0.27 | 6.75 | 8.92 | 79.89 | 88.94 | 8.51 | 16.43 |
| 7 | 6 | b | 0.62 | 0.26 | 5.53 | 8.44 | 90.21 | 102.45 | 13.75 | 15.90 |
| 8 | 7 | b | 0.54 | 0.25 | 5.98 | 7.99 | 86.94 | 99.64 | 11.86 | 19.76 |
| 9 | 7 | b | 0.71 | 0.23 | 7.32 | 10.32 | 79.73 | 112.64 | 9.86 | 18.43 |
| 10 | 9 | b | 0.65 | 0.32 | 6.88 | 9.66 | 85.87 | 107.93 | 13.72 | 21.67 |
| 11 | 10 | с | 0.46 | 0.39 | 8.71 | 11.05 | 108.12 | 123.54 | 12.89 | 19.67 |
| 12 | 10 | с | 0.48 | 0.28 | 7.56 | 10.55 | 94.55 | 115.80 | 12.64 | 22.35 |
| 13 | 10 | с | 0.61 | 0.43 | 9.14 | 11.79 | 88.58 | 119.50 | 16.87 | 22.89 |
| 14 | 11 | с | 0.58 | 0.39 | 8.31 | 10.66 | 112.06 | 128.40 | 14.51 | 20.12 |
| 15 | 13 | с | 0.79 | 0.45 | 9.54 | 11.88 | 94.67 | 132.40 | 16.21 | 18.34 |
| 16 | 16 | d | 0.68 | 0.54 | 10.12 | 11.69 | 109.32 | 134.75 | 15.95 | 17.98 |
| 17 | 17 | d | 0.80 | 0.63 | 9.69 | 12.32 | 98.52 | 125.95 | 19.43 | 19.37 |
| 18 | 22 | d | 0.76 | 0.76 | 11.32 | 12.90 | 126.41 | 134.98 | 17.86 | 23.43 |

| 19 | 20 | e | 0.89 | 0.72 | 10.31 | 12.33 | 122.58 | 140.20 | 19.31 | 22.93 |
|----|----|---|------|------|-------|-------|--------|--------|-------|-------|
| 20 | 24 | e | 0.85 | 0.70 | 10.88 | 13.20 | 129.82 | 140.77 | 17.54 | 24.21 |
| 21 | 29 | e | 0.88 | 0.74 | 10.63 | 13.10 | 127.93 | 142.21 | 18.76 | 25.32 |

* Age: $a = \le 20 \text{ y}, b = 21-30 \text{ y}, c = 31-40 \text{ y}, d = 41-50 \text{ y}, e = 51-60 \text{ y}$

Table 3. Heavy metals levels ($\mu g/g$) in fingernails and scalp hair samples of auto welding workers

| No | Exposure duration, | Δœ | Cd | | Ni | | Zn | | Pb | |
|----|--------------------|-----|-------|-------|-------|-------|--------|--------|-------|-------|
| NO | Year(y) | Age | Nails | Hairs | Nails | Hairs | Nails | Hairs | Nails | Hairs |
| 1 | 1 | а | 0.37 | 0.39 | 8.89 | 9.45 | 83.65 | 90.43 | 8.21 | 8.97 |
| 2 | 1 | а | 0.43 | 0.42 | 7.94 | 8.86 | 99.64 | 88.71 | 9.71 | 7.82 |
| 3 | 3 | а | 0.35 | 0.39 | 8.61 | 8.21 | 91.63 | 112.23 | 7.10 | 9.63 |
| 4 | 5 | b | 0.39 | 0.46 | 7.89 | 10.43 | 113.7 | 126.91 | 9.42 | 7.96 |
| 5 | 5 | b | 0.46 | 0.46 | 9.71 | 9.23 | 94.95 | 107.68 | 8.75 | 10.55 |
| 6 | 7 | b | 0.38 | 0.49 | 10.43 | 9.78 | 89.91 | 99.65 | 9.07 | 12.43 |
| 7 | 10 | b | 0.47 | 0.43 | 8.57 | 8.97 | 99.72 | 130.66 | 12.15 | 11.85 |
| 8 | 11 | b | 0.43 | 0.40 | 8.94 | 9.97 | 114.71 | 125.56 | 10.13 | 13.73 |
| 9 | 13 | с | 0.59 | 0.47 | 11.89 | 8.95 | 98.95 | 114.83 | 9.54 | 15.74 |
| 10 | 14 | с | 0.39 | 0.54 | 10.75 | 10.55 | 105.87 | 109.59 | 13.81 | 14.86 |
| 11 | 14 | c | 0.47 | 0.51 | 12.70 | 11.23 | 112.80 | 125.65 | 12.43 | 15.97 |
| 12 | 17 | с | 0.55 | 0.46 | 9.95 | 10.65 | 98.86 | 114.84 | 9.96 | 14.85 |
| 13 | 18 | с | 0.47 | 0.58 | 13.71 | 12.16 | 96.06 | 132.88 | 14.83 | 16.72 |
| 14 | 20 | с | 0.46 | 0.48 | 10.86 | 11.85 | 122.75 | 140.32 | 12.66 | 16.79 |
| 15 | 20 | с | 0.57 | 0.54 | 12.08 | 12.76 | 102.24 | 134.86 | 16.70 | 15.55 |
| 16 | 21 | с | 0.63 | 0.58 | 14.06 | 13.53 | 90.98 | 156.98 | 15.12 | 17.89 |
| 17 | 20 | d | 0.51 | 0.58 | 13.81 | 11.92 | 141.81 | 149.98 | 15.79 | 17.43 |
| 18 | 24 | d | 0.62 | 0.65 | 12.95 | 13.27 | 138.9 | 161.21 | 17.54 | 18.89 |
| 19 | 25 | d | 0.58 | 0.61 | 14.00 | 13.08 | 150.70 | 155.90 | 15.85 | 18.52 |
| 20 | 28 | e | 0.72 | 0.63 | 13.75 | 13.14 | 147.90 | 160.13 | 16.75 | 19.21 |

* Age: a = \leq 20 y, b = 21-30 y, c = 31-40 y, d = 41-50 y, e = 51-60 y

The data of unpolluted workplace sites (Table 1) and autoworkers (Tables 2-5) were compared. In general, it was noted that the concentrations of toxic and heavy metals were higher in autoworkers samples than that in control samples, and this is in agreement with that reported by others for similar studies (Al-Easawi et al., 2017; Wang et al., 2009). The impact of age and exposure duration per year on element concentrations was investigated. It was found that clear relation between the concentrations of toxic and heavy metals influence age and exposure duration. Consequently, many factors contributed to introducing toxic and heavy metals into biological systems; this means the term of tolerant levels suitable for use instead of reference values (Pozebon et al., 2017; Alrobaian & Arida, 2019; Lotah et al., 2022). In this context, the average levels of Cd were found to be slightly increased upon repair car tires workers (Table 4) compared with other autoworkers (Tables 2, 3 and 5). On the other hand, the concentrations of Zn in auto welding workers (Table 3) were found to be significantly higher compared with that in other autoworkers (Tables 2, 4 and 5). The concentration average levels of Pb and Ni in the washing and oil change services center workers' fingernails and scalp hair volunteers (Table 5) were found to be slightly increased compared with other

autoworkers (Tables 2-4). A comparison of the toxic and heavy elements' concentrations in fingernails and scalp hair of autoworkers and control volunteers was carried out by using Microsoft Excel of Office 365 to calculate the T-test. It was determined that didn't normal distribution, a value p < 0.05 was considered to be significant. These results are consistent with other similar studies as well as below the international tolerance levels (Gönener et al., 2020; Nakaona et al., 2020). As a result of this study, a statistically significant relationship has been found between autoworkers and the toxic and heavy metal levels in the fingernails and scalp hair of volunteers. Indeed, once a toxic and heavy element is absorbed, it distributes in tissues and organs, later on, excretion typically occurs primarily through kidneys and digestive tract, some elements tend to persist in some storage sites, like the liver, bones, hair, and kidneys, for many years (Pozebon et al., 2017; Alrobaian & Arida, 2019; Lotah et al., 2022). In general, the sequences for the concentration of investigated metals in groups (unpolluted workplace site (control), car mechanics workers, car mechanics workers, and washing and oil change services center workers) are the following: Zn> Pb > Ni >Cd, as shown in (Tables 1-3 and 5) while the repair car tires workers are $Zn > Ni \ge Pb > Cd$, as shown in **Table 4**. As a result of this research,

it has been revealed that there is a statistically significant relationship between environmental including the effects of the workplace of autoworkers, and the toxic and heavy metal levels detected in their fingernails and scalp hair.

Table 4. Heavy metals levels $(\mu g/g)$ in fingernails and scalp hair samples of repair car tires workers

| No | Exposure duration, | A | Cd | | N | i | 2 | Zn | Pb | |
|----|--------------------|----------|-------|-------|-------|-------|--------|--------|-------|-------|
| NO | Year(y) | Age | Nails | Hairs | Nails | Hairs | Nails | Hairs | Nails | Hairs |
| 1 | 2 | а | 0.38 | 0.45 | 9.15 | 8.79 | 70.14 | 69.37 | 6.98 | 7.20 |
| 2 | 2 | а | 0.43 | 0.51 | 10.12 | 9.43 | 61.5 | 71.22 | 8.43 | 6.54 |
| 3 | 5 | b | 0.58 | 0.41 | 11.62 | 8.95 | 83,52 | 66.21 | 8.98 | 8.41 |
| 4 | 7 | b | 0.61 | 0.64 | 9.20 | 10.68 | 98.72 | 81.80 | 11.21 | 11.25 |
| 5 | 7 | b | 0.55 | 0.59 | 12.61 | 9.87 | 85.74 | 79.77 | 10.95 | 10.55 |
| 6 | 8 | b | 0.64 | 0.68 | 12.84 | 12.33 | 78.91 | 91.87 | 11.79 | 11.4 |
| 7 | 10 | b | 0.49 | 0.73 | 10.77 | 12.82 | 89.63 | 94.71 | 9.75 | 10.6 |
| 8 | 11 | b | 0.61 | 0.68 | 14.08 | 11.85 | 95.11 | 98.70 | 13.06 | 11.1 |
| 9 | 11 | b | 0.40 | 0.72 | 11.83 | 13.22 | 90.66 | 91.85 | 8.89 | 9.69 |
| 10 | 13 | b | 0.54 | 0.79 | 13.70 | 12.14 | 113.70 | 99.62 | 12.85 | 11.8 |
| 11 | 12 | с | 0.71 | 0.72 | 11.72 | 11.91 | 102.45 | 105.53 | 10.76 | 13.8 |
| 12 | 18 | с | 0.49 | 0.88 | 13.05 | 13.76 | 88.69 | 108.80 | 9.63 | 12.8 |
| 13 | 19 | с | 0.68 | 0.82 | 12.92 | 12.60 | 90.63 | 98.97 | 11.85 | 12.5 |
| 14 | 19 | с | 0.57 | 0.78 | 15.48 | 14.81 | 104.76 | 113.23 | 14.21 | 14.1 |
| 15 | 20 | с | 0.74 | 0.85 | 13.38 | 13.93 | 98.06 | 106.60 | 12.09 | 13.6 |
| 16 | 22 | с | 0.64 | 0.84 | 13.87 | 14.11 | 109.66 | 114.85 | 14,94 | 11.7 |
| 17 | 23 | с | 0.80 | 0.78 | 15.47 | 15.21 | 112.46 | 119.65 | 14,87 | 14.8 |
| 18 | 21 | d | 0.64 | 0.79 | 14.50 | 14.47 | 99.64 | 113.82 | 16.86 | 12.9 |
| 19 | 26 | d | 0.77 | 0.77 | 13.76 | 15.48 | 119.58 | 121.85 | 16,65 | 13.6 |
| 20 | 30 | d | 0.69 | 0.91 | 14.20 | 16.67 | 119.61 | 126.31 | 15.08 | 15.9 |
| 21 | 29 | e | 0.72 | 0.88 | 13.21 | 15.93 | 109.60 | 122.89 | 13.93 | 15.2 |
| 22 | 31 | e | 0.78 | 0.84 | 15.96 | 16.88 | 113.87 | 126.51 | 14.08 | 14.7 |

* Age: $a = \le 20$ y, b = 21-30 y, c = 31-40 y, d = 41-50 y, e = 51-60 y

Table 5. Heavy metals levels ($\mu g/g$) in fingernails and scalp hair samples of washing and oil change services center workers

| No | Exposure duration, | 1 00 | C | d | Ν | Ni | Z | n | Pb | |
|----|--------------------|------|-------|-------|-------|-------|--------|--------|-------|-------|
| NO | Year(y) | Age | Nails | Hairs | Nails | Hairs | Nails | Hairs | Nails | Hairs |
| 1 | 3 | а | 0.53 | 0.49 | 6.08 | 6.64 | 80.06 | 75.81 | 7.98 | 8.08 |
| 2 | 3 | b | 0.48 | 0.48 | 7.35 | 6.32 | 72.6 | 77.23 | 8.63 | 9.11 |
| 3 | 2 | b | 0.50 | 0.57 | 5.43 | 6.77 | 81.16 | 77.43 | 10.64 | 8.32 |
| 4 | 4 | b | 0.48 | 0.49 | 7.75 | 6.94 | 79.95 | 82.32 | 9.72 | 8.87 |
| 5 | 7 | b | 0.43 | 0.62 | 8.53 | 8.21 | 88.65 | 88.64 | 10.61 | 8.62 |
| 6 | 8 | b | 0.51 | 0.59 | 7.84 | 7.93 | 97.43 | 94.72 | 7.88 | 12.2 |
| 7 | 10 | b | 0.48 | 0.54 | 9.07 | 10.21 | 90.66 | 112.85 | 8.91 | 14.7 |
| 8 | 10 | b | 0.56 | 0.68 | 14.57 | 9.88 | 112.71 | 99.53 | 12.14 | 14.5 |
| 9 | 11 | b | 0.47 | 0.63 | 12.73 | 12.32 | 105.32 | 116.64 | 9.05 | 15.8 |
| 10 | 12 | b | 0.61 | 0.7.2 | 10.70 | 13.21 | 86.94 | 120.21 | 11.09 | 16.8 |
| 11 | 12 | b | 0.49 | 0.74 | 8.59 | 12.89 | 99.06 | 112.62 | 9.74 | 18.6 |
| 12 | 13 | b | 0.54 | 0.62 | 13.82 | 14.11 | 92.94 | 118.51 | 13.73 | 17.6 |
| 13 | 9 | с | 0.63 | 0.77 | 10.84 | 13.54 | 131.71 | 108.98 | 11.82 | 15.6 |
| 14 | 12 | с | 0.54 | 0.67 | 15.82 | 15.88 | 112.61 | 116.73 | 13.79 | 12.3 |
| 15 | 13 | с | 0.61 | 0.65 | 17.95 | 18.79 | 98.79 | 124.71 | 12.77 | 17.1 |
| 16 | 13 | с | 0.56 | 0.74 | 17.09 | 16.76 | 133.71 | 121.43 | 14.81 | 16.8 |

| 17 | 21 | d | 0.71 | 0.82 | 13.85 | 15.89 | 116.86 | 129.64 | 14.89 | 19.21 |
|----|----|---|------|------|-------|-------|--------|--------|-------|-------|
| 18 | 18 | d | 0.64 | 0.79 | 14.71 | 16.84 | 136.82 | 133.62 | 17.63 | 18.53 |
| 19 | 19 | d | 0.70 | 0.85 | 15.80 | 17.54 | 144.65 | 129.66 | 16.88 | 20.21 |
| 20 | 28 | e | 0.60 | 0.87 | 17.73 | 18.54 | 124.70 | 130.41 | 18.76 | 20.14 |

* Age: $a = \le 20 \text{ y}, b = 21-30 \text{ y}, c = 31-40 \text{ y}, d = 41-50 \text{ y}, e = 51-60 \text{ y}$

Conclusion

The levels of certain toxic and heavy metals (Pb, Zn, Cd, and Ni) were determined by using ICP-MS in fingernails and scalp hair of autoworkers in Saudi Arabia to study exposure to chemical pollutants metals. It was found that the concentrations of toxic and heavy metals were higher in autoworkers samples than that in control samples Toxic and heavy metals levels increase with increase the age years of autoworkers, epically for Pb and Cd. The obtained result proved that fingernails and scalp hair could be used as a good biological indicator for the assessment of toxic heavy metal pollution. On the other hand, this research shows that there is a statistically significant relationship between the type of work (autoworkers) or workplace and the heavy metal concentration detected in fingernails and scalp hair samples, which represent a very important guide for the labor and health organizations.

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