Assessment of heavy metal levels in blood of metal fabricating factory workers in Nnewi, Nigeria

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Abstract
Introduction of Study: Heavy metal contamination or toxicity poses serious deleterious effects to human health.

Materials and Methods: This is a cross sectional study designed to assess the heavy metal levels in blood of metal fabricating factory workers in Nnewi. A total of 39 apparently healthy individuals in the exposed group (metal fabricating factory workers) aged between 19 and 56 years and 79 control individuals (comprising of 39 control individuals from Nnewi (N) and 40 control individuals from Elele (E) respectively) aged between 18 and 44 years were recruited for the study. Demographic data and anthropometric indices (BMI) of the study participants were obtained using structured questionnaire and thereafter, 10ml of venous blood sample was collected from each individual for the evaluation of heavy metal levels (Pb, Ni, Cu, Zn, As and Se) using atomic absorption spectroscopy (AAS). Results: This study showed that these workers had an average length of service (LOS) of 11.92±2.77 years with no significant difference in body mass index (BMI) of factory workers compared with control N and E individuals (p>0.05) respectively. Ni, As and Pb levels and Cu, Zn and Se levels were significantly elevated and reduced respectively in factory workers than in controls (p<0.05). Metal levels differed significantly with both age and length of service (LOS) in exposed worker compared with control individuals (p<0.05). However, no significant statistical correlations were found between metal levels in factory workers and age as well as LOS (p>0.05).

Conclusion: This study showed elevated levels of Ni, As and Pb and decreased levels of Cu, Zn and Se in the blood of metal fabricating factory workers and this demands urgent attention.

Keywords: Metal fabricating factory, Factory workers, Heavy metals, Body mass index (BMI), Age, Length of service (LOS).

Introduction
Metal fabrication is the building of metal structures by cutting, bending, and assembling processes. It is a value added process that involves the creation of machines, parts, and structures from various raw materials.¹ As with other manufacturing processes, both human labour and automation are commonly used.

Heavy metals are natural elements characterized by their rather high atomic mass and their high density. Although typically occurring in rather low concentration, they can be found all through the crust of our planet.² Some heavy metals like copper, selenium, or zinc are essential trace elements, with functions indispensable for various biological processes also driving the entire human metabolism. For instance, the heavy metal cobalt, acting as the central atom in the vitamin B12 complex, is a key player in the reductive branch of the propionic acid fermentation pathway³ and without this special heavy metal compound, the gourmet for instance would have to do without the unique flavor of Emmentaler cheese. Nevertheless, many of them, e.g., mercury, cadmium, arsenic, chromium, thallium, lead, and others, classically represent the “dark side of chemistry”⁴ in that they exert toxic effects even at low concentrations.⁵

Heavy metals are significant environmental pollutants and their toxicity is a problem of increasing significance for ecological, evolutionary, nutritional and environmental reasons.⁶ The toxicity of heavy metals depends on a number of factors. Specific clinical manifestations vary according to the metal in question, the total dose absorbed, and whether the exposure was acute or chronic.⁷ The age of the person can also influence toxicity. For example, young children are more susceptible to the effects of lead exposure because they absorb several times the percent ingested compared with adults and because their brains are more plastic and even brief exposures may influence developmental processes.⁸ The route of exposure is also important e.g elemental mercury is relatively inert in the gastrointestinal tract and also poorly absorbed through intact skin, yet inhaled or injected elemental mercury may have disastrous effects.⁹ Heavy metals are significantly related with the incidence of acute and chronic disorders, not only do they play an essential role in disrupting homeostasis of biochemical reactions in human body, but also, through their dissemination and storage in different tissues.¹⁰ The concentration of trace metals may vary from tissue to tissue throughout the body, for example metals concentration is higher in liver than the blood stream¹¹. Environmental factors like temperature, pH, water hardness, and organic matter can influence the toxicity of metals in biological systems.¹² Exposure to metals can occur through variety of routes and they may be inhaled as dust of fume or smoking, or even ingested involuntarily through food and drink. Bioaccumulation of these heavy metals can lead to
various disorders and can also result in excessive damage due to oxidative stress induced by free radical formation.\textsuperscript{10}

Ahmed\textsuperscript{9} in his study observed that heavy metals such as lead and zinc deposition in blood tissues showed a dramatic increase in relation with both age and length of exposure. A study carried out by Muhammad\textsuperscript{11} recorded significantly higher concentrations of toxic metals in exposed workers. Also, Ahed \textit{et al.}\textsuperscript{12} in their study involving 47 male workers with average mean age of 31.7 years recorded higher levels of copper and nickel in exposed individuals than in control individuals. Therefore, this cross sectional study was designed to assess the heavy metal levels in the blood of metal fabricating factory workers in Nnewi, South eastern Nigeria.

\section*{Materials and Methods}

\subsection*{Inclusion Criteria}

Apparently healthy individuals aged between 19 and 56 years who are exposed to metal fabricating factory effluents and control individual (non-exposed groups) were included in this study.

\subsection*{Study Design and Participant Recruitment}

This is a cross-sectional study designed to evaluate some heavy metal levels in blood of metal fabricating factory workers in Nnewi, Nigeria. A total of 15 apparently healthy individuals in the exposed group (metal fabricating factory workers) aged between 19 and 56 years were recruited for the study. The exposed group comprised workers from metal fabricating factory who were constantly being exposed to effluents from the factory. The control groups were made up of two (2) sets: The first set was made up of thirty-nine (39) staff and undergraduate students of the College of Health Sciences, Nnamdi Azikiwe University, Nnewi Campus whose residential homes were at least 5-10 km from the factory sites, while the second set was made up of forty (40) staff and undergraduate students of the Faculty of Medicine, Madonna University, Elele. They were aged between 18 and 44 years. Informed consent was obtained from all individuals after being educated on the benefit of the study and completing of a structured questionnaire. Thereafter, 10ml of venous blood sample was collected from each individual for the evaluation of heavy metal levels. Blood samples for the determination of lead (5ml) were delivered into new EDTA containers, mixed and stored frozen at -20°C until analyzed. The rest of the blood sample was delivered into lithium heparin containers and then centrifuged for 3 minutes at 2000 rpm. The plasma were separated and put into clean dry sample containers and stored deep-frozen at -4°C until analyzed. The plasma was used for the estimation of heavy metals (Pb, Ni, Cu, Zn, As and Se) by atomic absorption spectroscopy (AAS) according to the method of Smith \textit{et al.}\textsuperscript{13} Determination of lead in whole blood was done using the method as described by Hessel.\textsuperscript{14}

\subsection*{Exclusion Criteria}

Individuals of any known kidney disease, liver disorder, alcoholics and smokers as well as those outside the age limits were excluded from the study.

\section*{Ethical Consideration}

Ethical approval for the research was obtained from Ethical Committee, Nnamdi Azikiwe University Teaching Hospital, Nnewi, Anambra State, Nigeria (NAUTH/CS/66/Vol.2/149).

\section*{Statistical Analysis}

The data were presented as mean±SEM and the mean values of the control and test group were compared by Student’s t-test and Pearson’s bivariate correlation coefficient using Statistical package for social sciences (SPSS) (Version 16) software. A P<0.05 was considered as significant.

\section*{Results}

Table 1 shows the demographic profile of factory workers which captured the ages, heights, weights, BMIs of control individuals and workers in metal fabricating factory and the length of service of those workers. From the table, there was a significant mean difference between ages of control individuals from both Nnewi (N) and Elele (E) when compared with the ages of the metal fabricating factory workers (p<0.05). The mean age of the control individuals N and E did not differ significantly (p>0.05). There was a significant difference between the mean weight of control individuals from Nnewi when compared with the factory workers (p<0.05). Conversely, no significant difference in mean weight was noted in control individuals from Elele when compared with those of the factory workers (p>0.05). Furthermore, no significant difference was observed between the mean weight of control individuals from Nnewi and those from Elele although a higher mean weight was seen in control subjects from Nnewi compared to that in control individuals from Elele. Interestingly, no mean difference in BMI existed between the control individuals (both Nnewi and Elele) when compared with factory workers (p>0.05). Meanwhile, higher mean BMI in the control groups was observed when compared with the factory workers although the difference is not significant (p>0.05). Also, these workers had an average length of service (LOS) of 11.92±2.77 years.

The mean serum levels of Ni, Cu, Zn, As and Se and whole blood Pb of controls N and E subjects and factory workers are presented in Table 2. The levels of Cu, Zn, As, Se and Pb levels from control E individuals were higher and statistically different (p<0.05) from those of control N individuals, however, no significant difference (p>0.05) was observed for Ni.

The Ni level of metal fabricating (3.30±0.09) factory workers was highly elevated and statistically different (p<0.05) when compared to control N individuals (0.04±0.00) and control E individuals (0.07±0.00). The Cu level in the metal fabricating factory workers (8.91±0.30) was significantly reduced (p<0.05) when compared with control N (16.69±0.21) and control E (19.72±0.21) individuals. Zn levels of the factory workers of 5.75±0.41 were statistically reduced (p<0.05) when compared with the controls N (11.73±0.19) and E (17.11±0.46) individuals. However, the As level in metal fabricating factory workers was significantly elevated compared to both controls N and E individuals (p<0.05). Serum Se levels were reduced in the
factory workers when compared with the control (p<0.05), whereas, the Pb level in the metal fabricating factory workers were significantly elevated when compared with control N (0.59±0.01) and E (0.79±0.10) individuals (p<0.05).

The effects of age on the metal levels of metal fabricating factory workers are presented in Table 3 while Fig. 1 presents the regression coefficients of metal levels with age. There were significant elevations (p<0.05) in Ni, As and Pb levels and significant reduction (p<0.05) in Cu, Zn and Se levels compared to both control N and E individuals in all the age groups. Ni, Cu and Zn were positively correlated with age with regression coefficients (r) of 0.410, 0.121 and 0.063, respectively while As, Se and Pb were negatively correlated with age with r values of -0.381, -0.208 and -0.138, respectively.

Table 4 presents the effect of LOS on the metal levels of the metal fabricating factory workers while Fig. 2 presents the regression analyses with LOS. There were significant increases (p<0.05) in Ni, As and Pb levels in all the LOS groups as against the control while Cu, Zn and Se levels were significantly reduced (p<0.05) compared with both controls N and E individuals. The highest metal level were obtained at the 16-20yr group for Ni and As; and 6-10yr group for Pb. Nickel (r=0.335; p=0.218) and Cu (r=0.113; p=0.690) were positively correlated with LOS while Zn (r=-0.139; p=0.624), As (r=-0.606; p=0.832), Se (r=-0.355; p=0.192), and Pb (r=-0.147; p=0.601) were negatively correlated with LOS, though non-significantly (p>0.05).

### Table 1: Demographic profile of metal fabricating factory workers

| Variables | Age (years) | LOS (years) | Weight (kg) | Height (m) | BMI (Kg/m²) |
|-----------|-------------|-------------|-------------|-----------|-------------|
| N (n=39)  | 23.2±0.91   | 74.82±0.04  | 1.74±0.01   | 24.7±0.38 |
| E(n=40)   | 21.6±0.33   | 66.10±1.09  | 1.68±0.01   | 23.58±0.67 |
| M (n=15)  | 37.8±3.15   | 65.73±2.79  | 1.68±0.02   | 23.37±1.06 |

*Values are in mean (±SEM); within the column, means with different superscripts are statistically significant (p<0.05). Key:
N: Control individuals from Nnewi
E: Control individuals from Elele
M: Workers from metal fabricating factory
BMI: Body mass index
LOS: Length of service

### Table 2: Metals levels in metal fabricating factory workers

| Variables | Ni (µmol/L) | Cu (µmol/L) | Zn (µmol/L) | As (µmol/L) | Se (µmol/L) | Pb (µmol/L) |
|-----------|-------------|-------------|-------------|-------------|-------------|-------------|
| N(n=39)   | 0.04±0.00   | 16.69±0.21  | 11.73±0.19  | 0.01±0.00   | 5.11±0.08   | 0.59±0.01   |
| E(n=40)   | 0.07±0.00   | 19.72±0.21  | 17.11±0.46  | 0.02±0.00   | 6.66±0.08   | 0.79±0.10   |
| M(n=15)   | 3.30±0.09b  | 8.91±0.30bc | 5.75±0.41f  | 0.07±0.00b  | 3.63±0.10e  | 1.11±0.09c  |

*Values are in mean (±SEM); within the column, means with different superscripts are statistically significant (p<0.05). Key:
N: Control individuals from Nnewi
E: Control individuals from Elele
M: Workers from metal fabricating factory

### Table 3: Effect of age on heavy metals levels of metal fabricating factory workers

| Age group | Ni (µmol/L) | Cu (µmol/L) | Zn (µmol/L) | As (µmol/L) | Se (µmol/L) | Pb (µmol/L) |
|-----------|-------------|-------------|-------------|-------------|-------------|-------------|
| Controls (n=39) | 0.04±0.00a  | 16.69±0.21a | 11.73±0.19b | 0.01±0.00a  | 5.11±0.08b  | 0.59±0.01a  |
| 18-30yrs (n=3)  | 3.24±0.17bc | 8.74±0.36a  | 6.03±0.67a  | 0.70±0.00a  | 3.70±0.13a  | 1.13±0.05b  |
| 31-40yrs (n=3)  | 3.04±0.16bc | 8.41±0.22b  | 4.79±0.09a  | 0.08±0.00a  | 3.71±0.14a  | 1.11±0.05b  |
| 41-50yrs (n=5)  | 3.46±0.16ad | 9.72±0.75a  | 6.70±0.90a  | 0.07±0.01c  | 3.57±0.29a  | 1.08±0.05c  |
| 51-60yrs (n=2)  | 3.53±0.09ad | 8.24±0.26a  | 5.65±1.64a  | 0.50±0.00a  | 3.64±0.18a  | 1.14±0.03b  |

*Values are in mean (±SEM); within column, means with different superscripts are statistically significant (p<0.05).

### Table 4: Effect of LOS on heavy metal levels of metal fabricating factory workers

| LOS group | Ni (µmol/L) | Cu (µmol/L) | Zn (µmol/L) | As (µmol/L) | Se (µmol/L) | Pb (µmol/L) |
|-----------|-------------|-------------|-------------|-------------|-------------|-------------|
| Controls (n=39) | 0.04±0.00a  | 16.69±0.21b | 11.73±0.19b | 0.01±0.00a  | 5.11±0.08b  | 0.59±0.01a  |
| 0-5yrs (n=6)    | 3.16±0.15b  | 8.97±0.27a  | 5.71±0.55ab | 0.07±0.00b  | 3.79±0.11a  | 1.10±0.04b  |
| 6-10yrs (n=2)   | 3.34±0.09bc | 7.06±0.24ab | 7.06±0.24ab | 0.07±0.02ab | 3.61±0.18ab | 1.13±0.03ab |
| 11-15yrs (n=3)  | 3.23±0.22bc | 4.80±0.88a  | 4.80±0.88a  | 0.07±0.01bc | 3.46±0.18a  | 1.09±0.04b  |
| 16-20yrs (n=4)  | 3.41±0.14bc | 9.15±0.59a  | 5.41±0.72bc | 0.07±0.01bc | 3.50±0.19a  | 1.10±0.04b  |

*Values are in mean (±SEM); within column, means with different superscripts are statistically significant (p<0.05)

KEY:
LOS: Length of service
Fig. 1: Regression of metal levels of metal fabricating factory workers with age

Fig. 2: Regression of metal levels of metal fabricating factory workers with LOS
Discussion

Heavy metals with adverse health effects in human metabolism present obvious concerns due to their persistence in the environment.15 Both acute and chronic heavy metal intoxications and exposures have been implicated in the several degenerative conditions16 and may also increase the risk of some cancers.17

In this study, the mean levels of Cu, Zn, As, Se and Pb levels from control E individuals were higher and statistically different compared with control N individuals, but, no significant difference was observed for Ni. This may be due to the fact that Elele is a rural area where their predominant foods are protein-rich foods such as meat, poultry, fish and sea foods that are known to contain high levels of these metals.18 Another possible reason for this finding may be that Elele control individuals are living in the South-South zone in Nigeria where a lot of burning of fossil fuel such as flaring of gas whose deposits includes heavy metals settles on vegetation and food consumed by the people.19

The present study revealed that Ni level of metal fabricating (3.30±0.09) factory workers was significantly elevated when compared to control N and E individuals. This is in consonance with the report of El-Shafei20 who showed that 25 nickel-plating workers overwhelmingly suffered from compromised liver function due to high Ni level obtained in their blood. This result may be attributable to the prevalent environmental pollution in the area under study.

However, there was a significant reduction in the mean level of copper (Cu) in the factory workers than in the control individuals. This result agrees with the finding of El -Safy et al.21 who documented low levels of copper among galvanization workers in the iron and steel industry. However, the present result is in contrast with some previous studies22-24 Essential metals are coactivators of several important enzymes, including antioxidant enzymes, and proteins which are necessary for health maintenance.25,26 Copper metal is a very important component of many enzymes especially those involved in redox cycling, mitochondrial respiration and iron absorption.27 However, low level of Cu observed in this study in Nnewi suggest that the workers may be at risk of developing certain diseases associated with depleted copper level or may be prone to some defects due to the alterations in metabolic functions mediated by copper.

Furthermore, the mean serum level of zinc (Zn) in the factory exposed individuals was significantly reduced compared with the control individuals. This may be due to inadequate intake by these individuals or due to the influence of lead (Pb) on Zn. This is in contrast with the work of Adejumo et al. who recorded an elevated level of zinc in auto mechanics, spray painters and battery recyclers compared with the non-exposed controls.23 Furthermore, Koranteng-Addo et al. showed higher level of Zinc in human scalp hair in occupationally exposed workers in Cape Coast, Ghana.28 This decrease in zinc level in the factory workers in the present study may hamper some essential metabolic roles which are mediated by zinc in human beings.

In this study, Selenium level was significantly reduced in the factory workers than in control individuals. The decreased selenium levels as obtained in this study may pre-dispose these workers to selenium-deficiency related diseases such as male infertility and Kashin-Beck disease, a type of osteoarthritis that occurs in certain low-selenium areas of China, Tibet and Siberia.29,30

However, there were significant elevations in the mean levels of both As and Pb in the factory workers compared with the control individuals respectively. This is in keeping with previous studies,31,32 who observed a significant difference in the mean level of Pb in workers compared to the control group. Also, previously Job et al. had earlier noted a higher mean level of Pb in pregnant women in Nnewi.31 The Pb level observed in the present study indicates that the factory workers may suffer deleterious effects of lead toxicity. Also, the arsenic levels obtained in this work are above the 1µg/L (0.01µmol/L) recommended as permissible level in blood of adults.33 This calls for a serious concern bearing in mind that arsenic exposure can cause death34 and induce oxidative injury35 in exposed individuals.

There were significant elevations in Ni, As and Pb levels and significant reduction in Cu, Zn and Se levels compared to both control individuals in all the age groups. Ni, Cu and Zn were positively correlated while As, Se and Pb was negatively correlated with age respectively although non-significantly. This is in keeping with the findings of Ayibike et al.36 who observed no statistical difference in Pb level for all age groups. Liang et al.37 found no correlation existed between the concentrations of heavy metals which they studied and age. Their finding is consistent with the work of Lim et al.38 However, the reports of Ibeto and Okoye39 (2010) and Ahed et al.40 are in contrast with our present finding.

Interestingly, there were significant increases in the mean Ni, As and Pb levels in all the LOS groups as against the control while Cu, Zn and Se levels were significantly reduced compared with both controls N and E individuals. The highest metal level was obtained at the 16-20yr group for Ni and As; and 6-10yr group for Pb. Furthermore, no significant correlation was observed between the heavy metals studied and LOS in this study. The length and duration of exposure of workers in a particular environment may affect the level of heavy metals in the blood of the workers.41 Therefore, continuous exposure to elevated levels of metals could have serious health implications.42

Conclusion

This study revealed that Ni, As and Pb levels and Cu, Zn and Se levels were significantly elevated and reduced respectively in factory workers than in non exposed individuals. Metal levels differed significantly with both age and length of service (LOS) in exposed worker compared with control individuals, but no significant statistical correlations were found between metal levels in factory workers and age as well. This finding has great implications to human health and hence, demands urgent attention.

Conflict of Interest: None.
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