Assessment of heavy metal levels in blood of metal forging factory workers in nnewi, Anambra state, Nigeria

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Abstract

Background: Heavy metal toxicity has proved to be a major threat to human health as several health risks are associated with it.

Materials and Methods: This is a cross sectional study designed to assess the heavy metal levels in blood of metal forging factory workers in Nnewi, Anambra State, Nigeria. A total of eighteen (18) apparently healthy subjects from the exposed group (metal forging factory) aged between 19 and 56 years and 79 control subjects (comprising of 39 control subjects from Nnewi (N) and 40 control from Elele (E) respectively) aged between 18 and 44 years were recruited for the study. The body mass index (BMI) and length of service (LOS) of the subjects were obtained using structured questionnaire and thereafter, 10ml of venous blood sample was collected from each individual for the assessment of heavy metal levels (Pb, Ni, Cu, Zn, As and Se) using atomic absorption spectroscopy (AAS).

Results: The result revealed that the body mass index (BMI) of the metal forging factory workers differed significantly compared with control N subjects (24.75 ± 0.38) and E subjects (23.58 ± 0.67) (p<0.05). Factory workers had a length of service (LOS) of 5.74 ± 1.21 years. There were significant increases in the mean levels of Ni, As and Pb and decreases in Cu, Zn and Se levels in the metal forging factory workers compared with control subjects (p<0.05) respectively. Metal levels differed significantly with both age and length of service (LOS) in exposed worker compared with control subjects (p<0.05). Also, Zn level was significantly correlated with LOS of metal forging factory workers (r=0.562; p=0.015).

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

Keywords: Heavy metal, Metal forging factory, Body mass index (BMI), Length of Service (LOS).

Introduction

Metal forging is defined as a metal working process in which the useful shape of metal work piece is obtained in solid state by compressive forces applied through the use of dies and tools ¹. Forging process is accomplished by hammering or pressing the metal. It is one of the oldest known metal working processes with its origin about some thousands of years back. Traditionally, forging was performed by a smith using hammer and anvil. Using hammer and anvil is a crude form of forging. In modern times, industrial forging is done either with presses or with hammers powered by compressed air, electricity, hydraulics or steam.¹

Heavy metals are naturally occurring elements that have a high atomic weight and a density at least five times greater than that of water.² Some debate exists as to exactly what constitutes a "heavy metal" and which elements should properly be classified as such. Some authors have based the definition on atomic weight, while others point to those metals with a specific gravity of greater than 4.0, or greater than 5.0. Most recently, the term "heavy metal" has been used as a general term for those metals and semimetals with potential human or environmental toxicity.³ Heavy metal toxicity has proved to be a major threat as several health risks are associated with it. These metals have hardly any biological role to play in the human body but on the contrary their toxic effect causes malfunctioning of the body system.⁴ Anthropogenic activities such as seen in metal forging factories contribute significantly to environmental contamination. These metals are systemic toxicants known to induce adverse health effects in humans, including cardiovascular diseases, developmental abnormalities, neurologic and neurobehavioral disorders, diabetes, hearing loss, hematologic and immunologic disorders, and various types of cancer.⁵ The main pathways of exposure include ingestion, inhalation, and dermal contact. The severity of adverse health effects is related to the type of heavy metal and its chemical form, and is also time and dose-dependent. In biological systems, heavy metals have been reported to affect cellular organelles and components such as cell membrane, mitochondria, lysosome, endoplasmic reticulum, nuclei, and some enzymes involved in metabolism, detoxification, and damage repair.⁶ Metal ions have been found to interact with cell components such as DNA and nuclear proteins, causing DNA damage and conformational changes that may lead to cell cycle modulation, carcinogenesis or apoptosis.⁷ These elements may sometimes act as pseudo elements of the body, interfering with the metabolic processes that occur in the body. Metal toxicity depends on the absorbed dose, its route as well as duration of exposure. These heavy metals bind with protein sites which are not meant for them, by displacing original metals from their natural binding sites causing malfunctioning of cells by their toxicity.⁴ These heavy metals include; Cadmium, zinc, copper, lead, nickel, cobalt, arsenic, selenium, mercury and so on. These heavy metals, when they accumulate in the human body result in toxicity which causes several systemic health conditions especially in workers who are by their occupation exposed to them.

Maria *et al.* observed higher levels of heavy metals especially Pb in metal factory workers⁸. Muhammad also had a similar finding in which he observed significantly higher concentrations of toxic metals in the blood, urine, and saliva samples of exposed workers than in the control subjects⁹. The finding of Draz *et al.*¹⁰ was also in keeping with the observations of Maria *et al.*⁸ and Muhammed.⁹ However, Draz and his team observed no significant difference in blood metal levels in the different age groups and between both sexes in the factory workers group.¹⁰ Therefore, this study assessed the heavy metal levels in blood of metal forging factory workers in Nnewi, Anambra State, Nigeria.

Materials and Methods Study Design and Participant Recruitment

This is a cross-sectional study designed to assess heavy metal levels in blood of metal forging factory workers in Nnewi, Anambra State, Nigeria. A total of eighteen (18) apparently healthy subjects in the exposed group (metal forging factory workers) aged between 19 and 56 years were recruited for the study. The exposed group comprised workers from metal forging 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 -4^{0} 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^{0} 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 *et al.*¹¹ Determination of lead in whole blood was done using the method as described by Hessel.¹²

Inclusion Criteria

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

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.

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).

Statistical Analysis

The data were presented as mean \pm SEM and the mean values of the control and test group were compared by Student's ttest and Pearson's bivariate correlation coefficient using Statistical package for social sciences (SPSS) (Version 16) software. A P<0.05 was considered as significant.

Results

Demographic Profiles

The metal forging factory had the length of service (LOS) of 5.74 ± 1.21 years. The age, weight and body mass index (BMI) of the metal forging factory workers differed significantly when compared with control N subjects (24.75\pm0.38) and E subjects (23.58\pm0.67) (p<0.05) respectively. See table 1.

Table 1: Some demographic profiles of metal forging factory workers

Factory	Age (yrs)	LOS (yrs)	Weight (kg)	Height (m)	BMI (kg/m ²)
N (n=39)	23.28±0.91 ^{ab}		$74.82 \pm 1.04^{\circ}$	1.74 ± 0.01^{b}	24.75 ± 0.38^{b}
E (n=40)	21.68±0.33 ^a		66.10±10.91 ^b	1.68±0.01 ^a	23.58±0.67 ^b
K (n=18)	26.17±1.45 ^b	5.74±1.21	58.00±1.92 ^a	1.65 ± 0.02^{a}	21.37±0.69 ^a

Values are in mean (\pm SEM); within the column, means with different superscripts are statistically significant (p<0.05). Key:

- N: Control subjects from Nnewi
- **E**: Control subjects from Elele
- **K**: Workers from metal forging factory

BMI: Body mass index

LOS: Length of service

Metal levels of control (N and E) subjects and metal forging factory workers

The mean Ni, As and Pb levels were significantly elevated in metal forging factory workers than in control N subjects (0.04 ± 0.00) and control E subjects (0.07 ± 0.00) , (p<0.05) respectively. However, the mean concentrations of Cu, Se and Zn were significantly reduced in the metal forging factory workers when compared with the control subjects respectively (p<0.05). See table 2.

Table 2: Metal levels of metal forging factory workers

Factory	Ni(µmol/L)	Cu (µmol/L)	Zn (µmol/L)	As (µmol/L)	Se (µmol/L)	Pb (µmol/L)
N(n=39)	$0.04{\pm}0.00^{a}$	16.69±0.21 ^c	11.73 ± 0.19^{d}	$0.01{\pm}0.00^{a}$	$5.11 \pm 0.08^{\circ}$	0.59 ± 0.07^{a}
E (n=40)	$0.07{\pm}0.00^{a}$	19.72 ± 0.21^{d}	17.11 ± 0.46^{e}	$0.02{\pm}0.00^{a}$	6.66 ± 0.08^{d}	0.79 ± 0.10^{b}
K (n=18)	2.56 ± 0.07^{b}	9.06±0.19 ^e	7.11±0.14 ^c	0.02 ± 0.00^{bc}	2.59 ± 0.17^{a}	$0.85 \pm 0.02^{\circ}$

Values are in mean \pm SEM; within the column, means with different superscripts are statistically significant (p<0.05). **Key:**

N: Control subjects from Nnewi

E: Control subjects from Elele

K: Workers from metal forging factory

The effects of age on metal levels of metal forging factory workers

The effects of age on metal levels of factory workers are presented in Table 3 while the regression analyses are presented in Figure 1. Ni, As and Pb levels in all the age groups were significantly elevated (p<0.05) compared with controls N and E subjects while Cu, Zn and Se levels were significantly reduced (p<0.05). Highest metal levels were obtained in the range 30-40yrs for Ni, As and Pb, however, reduced Zn and Se levels were obtained in the 30-40yrs while Cu in the 18-30yrs groups. The regression coefficients of Ni, As and Se of 0.083, 0.100 and 0.325, respectively showed that these metals were positively correlated with age while Cu (r=-0.012), Zn (r=-0.425) and Pb (r=-0.151) were negatively correlated.

Age group	Ni (µmol/L)	Cu (µmol/L)	Zn(µmol/L)	As (µmol/L)	Se (µmol/L)	Pb(µmol/L)
N (n=39)	$0.04{\pm}0.00^{a}$	16.69 ± 0.21^{b}	11.73 ± 0.19^{b}	0.01 ± 0.00^{a}	5.11 ± 0.08^{b}	0.59±0.01 ^a
18-30yrs	2.57 ± 0.09^{b}	9.17±0.21 ^a	7.18 ± 0.17^{a}	0.02 ± 0.00^{b}	2.71 ± 0.08^{a}	0.86 ± 0.02^{b}
(n=14)						
30-40yrs	2.66±0.11 ^b	8.11 ± 0.18^{a}	7.17 ± 0.40^{a}	0.02 ± 0.00^{b}	2.61 ± 0.07^{a}	0.89 ± 0.08^{b}
(n=2)						
40-50yrs	2.58 ± 0.17^{b}	9.14±0.04 ^a	7.10±0.01 ^a	0.02 ± 0.00^{b}	3.25 ± 0.58^{a}	0.81 ± 0.03^{b}
(n=2)						

Table 3: Effect of age on heavy metal levels of metal forging factory workers

Values are in mean (\pm SEM); within column, means with different superscripts are statistically significant (p<0.05) Key:

N: Control subjects

The effect of LOS on metal levels of metal forging factory workers

Table 4 presents the effect of LOS on metal levels of factory workers while Figure 2 presents the regression of metal levels with LOS. There were significant differences (p<0.05) between all the metal levels in all the LOS groups and the controls N and E subjects with elevated levels of Ni, As and Pb and reduced levels of Cu, Zn and Se. The highest levels of Ni and Pb were recorded in the 11-15yr group while As was in the 6-10yr group. Except for Zn (r=0.562; p=0.015) which was significantly correlated (p<0.05) with LOS, Ni (r=-0.371; p=0.130), Cu (r=-0.145; p=0.567), Se (r=-0.185; p=0.463) and Pb (r=-0.450; p=0.061) Ni, Cu, As, Se and Pb were negatively correlated but non-significantly (p>0.05) with LOS.



Fig. 1: Regression of metal levels of metal forging factory workers with age

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

LOSgroup	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^{a}	16.69±0.21 ^b	11.73±0.19 ^b	0.01 ± 0.00^{a}	5.11 ± 0.08^{b}	0.59±0.01 ^a
0-5yrs (n=12)	2.59±0.07°	9.09±0.24 ^a	6.93±0.15 ^a	0.02 ± 0.00^{b}	2.82±0.13 ^a	0.88 ± 0.02^{b}
6-10yrs (n=2)	2.75±0.20 ^{cd}	8.82±0.53 ^a	7.73±0.16 ^a	0.03 ± 0.01^{b}	2.61±0.07 ^a	0.80 ± 0.02^{b}
11-15yrs (n=2)	2.83 ± 0.09^{d}	9.22±0.12 ^a	7.33±0.24 ^a	0.02 ± 0.00^{b}	2.81 ± 0.14^{a}	0.89 ± 0.05^{b}
15-20yrs (n=2)	2.13±0.08 ^b	8.83 ± 0.74^{a}	7.91±0.26 ^a	0.02 ± 0.00^{b}	2.51 ± 0.01^{a}	0.78 ± 0.05^{b}

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

N: Control subjects

LOS: Length of service



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Effect of gender on factory workers' Metal levels

The effect of gender on the metal levels of the factory workers is presented in Table 5. There was no significant difference (p>0.05) between the Ni, Cu, Zn, As, Se and Pb levels of male and female workers of the metal forging factory.

Factories	Gender	Ni(µmol/L)	Cu(µmol/L)	Zn(µmol/L)	As(µmol/L)	Se(µmol/L)	Pb(µmol/L)
Ν	M(n=29)	0.37 ± 0.02^{a}	16.90 ± 0.24^{d}	11.85 ± 0.23^{d}	0.01 ± 0.00^{a}	$4.05\pm0.01^{\circ}$	0.59±0.01 ^a
	F (n=10)	0.38 ± 0.03^{a}	16.50 ± 0.48^{d}	11.06 ± 0.30^{d}	0.01 ± 0.00^{a}	$4.07 \pm 0.12^{\circ}$	0.59±0.03 ^a
Е	M(n=18)	0.08 ± 0.01^{a}	20.99 ± 0.35^{f}	16.29 ± 0.39^{e}	0.02 ± 0.00^{ab}	6.53 ± 0.13^{d}	$0.85 \pm 0.02^{\circ}$
	F (n=22)	0.05 ± 0.00^{a}	18.66 ± 0.36^{e}	17.79 ± 0.76^{f}	0.02 ± 0.00^{b}	6.76 ± 0.10^{d}	0.74 ± 0.02^{b}
Κ	M (n=8)	2.60 ± 0.09^{b}	8.91±0.30 ^{ab}	7.21 ± 0.24^{b}	0.02 ± 0.01^{b}	2.26 ± 0.32^{a}	$0.86 \pm 0.03^{\circ}$
	F (n=10)	2.51 ± 0.09^{b}	9.18±0.24 ^{ab}	7.14 ± 0.17^{ab}	0.02 ± 0.00^{b}	2.85 ± 0.14^{a}	$0.84 \pm 0.02^{\circ}$

Table 5: Effect of gender on metal levels of metal forging factory workers

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

KEY:

N: Control subjects from Nnewi

E: Control subjects from Elele

K: Workers from metal forging factory

M: Male

F: Female

Discussion

Heavy metals are significant environmental pollutants and their toxicity is a problem of increasing significance for ecological, evolutionary, nutritional and environmental reasons.¹³ Heavy metal toxicity has proven to be a major threat to human health with several health risks associated with it.¹⁴ The most commonly found heavy metals in waste water include arsenic, cadmium, chromium, and copper, lead, nickel, and zinc, all of which cause risks for human health and the environment ¹⁵. The present study investigated the heavy metal (Ni, Cu, Zn, As, Se and Pb) levels in the blood of metal forging factory workers in Nnewi town in Anambra State, South eastern Nigeria.

In this study, the mean age of the control subjects were (23.28 ± 0.91) and (21.68 ± 0.33) years for control N and E subjects respectively with the metal forging factory workers having a mean age of (26.17 ± 1.45) years. This shows that the studied subjects were from a young, active and vibrant population which forms the major part of the manufacturing sector of the economy. Further, the mean body mass index of the metal forging factory workers were significantly lower compared with the control subjects and had a mean length of service (LOS) of 5.74 ± 1.21 years.

Expectedly, the mean nickel (Ni) level was significantly elevated in the metal forging factory worker than in control subjects. This is in consonance with the previous report of Okpogba *et al.* who showed an elevated level of Ni in the blood of metal fabricating factory workers in Nnewi.¹⁶ This result may be as a result of the environmental pollution occurring in the area under study. Environmental nickel levels depend especially on natural sources, pollution from nickel-manufacturing industries and airborne particles from combustion of fossil fuels.¹⁷ Nickel is released into the atmosphere during nickel mining and the industrial

production of stainless steel and other nickel alloys, or by industries that use nickel and its compounds.¹⁸ The elevated level of Ni in this study may suggest that the exposed persons could suffer the effect of Ni toxicity. Previous report indicates that nickel is a potential immunomodulatory and immunotoxic agent aside from its action as an allergen in humans.¹⁹⁻²⁰

In the present study, the mean serum level of copper (Cu) was significantly reduced in metal forging factory workers than in control subjects. This is in keeping with previous studies.^{21,16} Copper is needed by the body for a number of functions, predominantly as a cofactor for a number of enzymes such as ceruloplasmin, cytochrome oxidase, dopamine β -hydroxylase, superoxide dismutase and tyrosinase.²² Therefore, depletion of Cu level may have negative implications for the affected persons.

Also, the mean serum levels zinc (Zn) and selenium (Se) was significantly reduced in metal forging factory workers than in control subjects. This is in contrast with the finding of Okpogba *et al.* on the heavy metal levels in chickens (*gallus gallus domestica*) in rural (Elele) and urban (Nnewi) areas.²³ This disparity in the result may be due to the fact that the birds were not exposed directly to the heavy metals as is in the case of the metal forging factory workers.

However, the mean levels lead (Pb) and arsenic (As) was significantly elevated in the metal forging factory workers compared to the control subjects. This is in agreement with the findings of some previous similar studies ^{24, 25, 16}. Also, Ziad *et al.* showed higher levels of Pb in Jordanian automobile workers which further agrees with the present result²⁶. The elevated levels of both lead and arsenic recorded in this study may imply that the exposed subjects in this study may experience lead and arsenic toxicities over time and hence, they are prone to suffering from the deleterious effects of both metals. Lead has been found to impair development and have harmful effects even at lower levels.^{27,28} Lead toxicity has been implicated as a potential cause of a wide range of multisystem defects or disorders in humans including impaired respiratory function,²⁹ infertility,³⁰ brain damage, renal defects and liver defects among others.³¹⁻³³ Recognizably, the most potent mechanism by which lead affects various systems of the human body is via its ability to induce oxidative stress by generating reactive oxygen species. On the other hand, As can induce cancer, genotoxicity and affect the hematopoietic

system, liver, kidneys, skin and brain ³⁴. Arsenic also exerts its toxicity by generating reactive oxygen intermediates during redox cycling and metabolic activation processes that cause lipid peroxidation and DNA damage.³⁵ Therefore, the elevated levels of lead and arsenic found in this study demands urgent health interventions.

Interestingly, Ni, As and Pb levels were significantly elevated while Cu, Se and Zn levels were reduced in all age groups than in control subjects respectively. No gender based difference was observed in metal levels studied in factory workers when compared with control subjects. However, Zn level was significantly correlated with length of service (LOS) of factory workers.

Conclusion

This study showed elevated levels of Nickel, Arsenic and Lead and decreased copper, Selenium and Zinc levels in metal forging factory workers than in control subjects respectively. There was no gender based differences observed in metal levels studied in factory workers when compared with control subjects. However, Zn level was significantly correlated with length of service (LOS) of factory workers (r=0.562; p=0.015).

Conflict of Interest: None.

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