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Welding fumes composition and their effects on blood heavy metals in albino rats

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Keywords: Bio-monitoring Heavy metals Toxicity Welding fumes	Toxic substances produced during welding include heavy metals, carbon monoxide, carbon dioxide, and nitrogen oxides. The study aims to evaluate the heavy metals concentration in welding fumes and the blood of the animals exposed to welding fumes. The fumes were collected from a welding site by a skilled welder and part of it was subjected to metals analysis. A total of 130 rats were divided into 13 groups. 12 groups were given doses calculated to correspond to real-life workers exposure regimes and 1 group served as control. The dosages were administered intratracheally after anesthetization weekly for 12 weeks. The animals were sacrificed and whole blood samples were collected for atomic absorption spectrophotometry. The metals in fumes analyzed were decreasing in order of Fe > K > Pb > Co > Cd > Ca > Ni > Mn > Zn > Cr > Al > Cu > Mg. Changes were observed in the behaviour of the test animals compared to the control indicating probable toxicity. The values of Pb, Cr, Fe, Mn, and Ni in the exposed animal's blood were higher than the control and increased relatively across the treatment groups. However, the values of Al and Zn were not significantly different from the control. These indicate that exposure to welding fumes having contained a significant amount of heavy metals has caused noticeable toxicity symptoms with simultaneous elevation in blood metal levels. Monitoring and regulation of these activities should be enforced by relevant authorities in Kano and Nigeria in general

1. Introduction

Welding is an industrial process that is widely performed and utilizes excessively higher temperatures to merge metals. Importantly, the process produces metallic fumes and gases that are regarded as potentially hazardous [1,2].

The biological hazards associated with welding fumes due to oxidation of components are well known [3]. Higher levels of welding fumes metals in the body result in adverse health conditions [4,5]. Wergeland and Iverson [6] cautioned that health risks may results from pneumonia associated with metal fumes exposure through welding, cutting, or grinding. Exposure to MS and SS welding fume has caused a mild systemic inflammatory response. The concentration of the particles from the breathing spaces correlated with the results inside the welding face shields [7]. Inhalation of galvanized spot welding fumes has resulted in acute lung toxicity largely due to the short-term exposure of particles that contain Zn [8].

Human health risks due to exposure to toxic metals are associated, as

a rule, with multiple factors. Such technologies as steel processing, electric arc welding, pyrometallurgy of heavy nonferrous metals, and electroplating bring about multimetallic pollution of the workroom and ambient air and other compartments of the environment including foodstuffs produced in contaminated areas [9].

The process of welding comprises the vaporization of an electrode or wire's metals and oxides which is used during the process to generate fumes or clouds of dust. Arc welding has been one of the most common types of welding, is the process of merging two pieces of metals that were transformed into liquid by heat when there is a passage of electricity from one electrical conductor to another [10]. During the process, fumes are generated at the tip of the electrode after the evaporation of metals and fluxes that coats the electrode. These metal vapors are condensed and oxidized after having contact with air and subsequently form small particulates that comprise a complex mixture of metal oxides [11]. The composition of welding fume in terms of elements is largely determined by the electrode composition and the material welded [12]. Excessive heat in welding operations is associated with high levels of

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fumes in industrial areas. The fumes were comprised of metal dust or metal oxide particles that have condensed from vapor [13]. The particles can easily be influenced by airflow and could distribute to places beyond the working area which are in turn absorbed by the welder's body [12]. The metal complexes that result, differ with the kind of metals/materials used and the process of welding used. As an example, stainless steel (SS) and mild-steel (MS), which form two of the most widely available kinds of electrode wire used, have different elemental compositions. SS fumes contain (Cr and Ni) which were found to be cytotoxic to human pulmonary cells and were related to lung diseases [14,15]. The penetration of all dust particles of Fe, Mn, and Si (nano and micro) into the human body often initiates a natural defense response, which does not only activate macrophages but also results in various inflammatory processes leading to some diseases [16–18].

In addition to heavy metals, other toxic substances released during the welding process include ozone, carbon monoxide, carbon dioxide, and nitrogen oxides. Production of Reactive Oxygen Species (ROS) is theorized as one of the mechanisms for acute adverse effects of welding fumes on health with stainless steels containing chrome and nickel producing more reactive oxygen species (ROS) than mild steel [19,20]. Therefore, assessment of effects by evaluation of heavy metals concentration in body fluids is mostly utilized as an exposure biomarker. Atomic Absorption Spectrometry (AAS) is a suitable process that monitors metals concentration during occupational exposure with a sensitive and rapid screening method [21]. There is a possibility of a long time of persistent release and exposure of workers to toxic metal pollutants may be at levels that damage tissue and organ in humans and other animals. The study would provide useful data required for setting standard protocols to limit exposure to metal fumes in urban Kano. The objective of the study was to determine the metals composition in welding fumes generated in the Kano metropolis and the concentration in the blood of experimental animals exposed to the fumes.

2. Materials and methods

2.1. Collection of welding fumes

The metal fumes used in the study were produced in a cubical open front fume chamber with a capacity of 1 m³. It was performed by a wellskilled welder performing manual metal welding (shielded manual metal arc welding) process that utilized a stainless steel hard surfacing electrode (Hyundai Welding electrode low hydrogen E 7018 3.2 mm) in Kofar Ruwa, Kano, and the fumes were subsequently collected on a 0.2µm nuclepore filters. They were collected in a significant amount at a specific welding site just before the start of the study. Only a single sample was collected for 1 h. The particle size of the collected welding fumes sample was evaluated by scanning electron microscopy (SEM). In addition, the fumes sample was suspended in distilled water and then sonicated for 1 min. The particle suspension (total sample) was incubated for 24 h at 37 °C. The suspensions of the sample were digested and then subjected to analysis by Atomic Absorption Spectrophotometer as described by Popstojanov et al. [22].

2.2. Experimental design

Albino rats were chosen for this study. The rats were obtained and housed at the Animal House, Department of Pharmacology, Aminu Kano Teaching Hospital, Kano, Nigeria. A randomized block design was adopted for this study. A total of 130 laboratory rats (*Rattus norvegicus*) were utilized for the study. The animals were maintained in the animal room and were allowed to acclimatize for two weeks before treatment. The animals weigh between 210–250 g. The animals were divided into 13 experimental groups with each group composed of 10 albino rats allocated randomly to the groups [23].

2.3. Housing and feeding conditions

The animal house was free of pathogens and other extraneous factors with restricted access. They were placed in cages with each cage housing 5 animals. The animals were marked on their tails with their respective dose ID for identification. The temperature of the animal room was maintained at about 22 °C (\pm 3 °C) and the relative humidity was at least 30 %. Concerning lighting, the pattern was 12 h light and 12 h dark. They were fed with a conventional laboratory diet and water *ad libitum*. There was adherence to the existing protocols for the use of lab animals strictly and ethical approval for the study was obtained from the College of Health Sciences Research Ethics Committee (CHS-REC), Bayero University, Kano [24].

2.4. Preparation of test substance

The study involved sub-chronic toxicity testing of the metal fumes in albino rats which lasted for 12 weeks and the treatment was administered to the animals weekly by intratracheal instillation [25]. The dosing paradigms used in the present study depict the real workplace exposures of metalworkers in Kano. A mathematical simulation was used to evaluate the daily lung burden of a metal worker over a specified number of hours work schedule and translated into appropriate doses administered to the rat [23,26,27]. Below are the endpoints and factors that were taken into account during the calculation:

- Fume concentration (5 mg/m, threshold limit value for welding fumes)
- Human minute ventilation volume (20,000 mL/min $\times 10^{-6}$ m³/mL)
- Exposure duration (no. of hr/day ×60 min/h)
- Deposition efficiency (15 %) [28,29].

Considering the above factors, metal workers daily burden for various hours per day

1 Metalworker daily burden (2 h/day) = Fume concentration (5 mg/m³) × Human minute ventilation volume (20,000 mL/min × $10-6m^3/mL$) × Exposure duration (2 h/day ×60 min/hr) × Deposition efficiency (15 %) = 1.8 mg

Using surface area of alveolar epithelium (rat = 0.4 m^2 ; human = 102 m^2) as dose metric [30]. Rat daily burden of exposure was taken as 0.0070 mg

Then, similar exposure in rats for 3yrs, 5yrs, 10yrs, and 20yrs will be 7.66 mg, 12.77 mg, 25.55 mg, and 51.10 mg respectively at 365 days per year. Each of these concentrations was then divided into 12 which was administered weekly for the period of the study (12 weeks)

- 2 Metalworker daily burden (4 h/day), As in above, similar exposure in rats for 3yrs, 5yrs, 10yrs, and 20yrs will be 15.44 mg, 25.73 mg, 51.46 mg, and 102.93 mg respectively at 365 days per year.
- 3 Metalworker daily burden (8 h/day), As in above, similar exposure in rat for 3yrs, 5yrs, 10yrs, and 20yrs will be 30.88 mg, 51.46 mg, 102.93 mg, and 205.86 mg respectively at 365 days per year [23].

Below are the working concentrations (dosage) of metal fumes administered on test animals for 12 weeks. Each concentration was given per animal per week [23].

Groups		
I	II	III
Group IA (0.64 mg/	Group IIA (1.29 mg/	Group IIIA (2.57 mg/
animal/week)	animal/week)	animal/week)
Group IB (1.06 mg/	Group IIB (2.14 mg/	Group IIIB (4.27 mg/
animal/week)	animal/week)	animal/week)
Group IC (2.13 mg/	Group IIC (4.29 mg/	Group IIIC (8.56 mg/
animal/week)	animal/week)	animal/week)
		(continued on next page)

(continued)

Group ID (4.26 mg/	Group IID (8.58 mg/	Group IIID (17.16 mg/
animal/week)	animal/week)	animal/week)

2.5. Administration and dose of test materials

Sterile saline was used to prepare the sample of the metal fumes and subsequently sonicated for 1 min to make the fumes dispersed. The rats were anesthetized with ketamine (0.1 mL/100 g b.w IP) and after passing out, immediately followed by intratracheal instillation of the respective dose per animal once a week for 12 weeks. However, 200 μ l of sterile saline was administered to the control animals through the intratracheal instillation was a commonly utilized technique to administer welding particulates into the lungs of laboratory animals. In such respect, welding fume is collected onto the filters, later suspended in an aqueous medium, and administered directly into the lungs of animals. Significance of such procedure over the inhalation technique includes simplicity, relatively low cost, and most importantly, the administration of a well-defined dose of particles [31–33].

2.6. Clinical symptoms

Within the 12 weeks dosing period, all the animals were observed daily for peculiar clinical signs and symptoms once before administration and immediately after for up to 3 h after dosing [34].

2.7. Collection of blood samples

Blood samples were collected 1-week post 12 weekly treatments. The animals were placed in a bucket containing wool soaked with chloroform to aid passing out. Samples were then collected from the jugular vein after cutting with a blade into an EDTA container for the analysis of heavy metals according to AVMA guidelines for euthanasia of animals [25,35].

2.8. Digestion of whole blood samples

Into about 2 mL of the blood sample which was stored previously at 4°C, concentrated nitric acid was added. Microwave is preferred for the preparation of the blood samples with involved rapid digestion. 1 mL of the whole blood sample was directly placed into a porcelain crucible. 3 mL of concentrated HNO3 -H2O2 (2:1, v/v) were added to the crucible. Pre-digestion was started by covering the crucible and keeping it at room temperature (~35 $^\circ\text{C})$ for about 5 min and then placing the crucibles in a microwave oven. One stage digestion was done after heating the crucibles which were at 30 % of total power (900 W). Digestion of the samples was terminated in 2-3 min after which the crucibles were allowed to cool down at room temperature and the resulting solution which comprise of about 0.5 mL of semi-dried mass was subsequently dissolved by 5 mL of 0.1 mol HNO3. They were then transferred quantitatively to a 10 mL volumetric flasks and followed by dilution with DDDW up to mark and transferred again to a polyethylene storage bottle for further analysis [36,37].

2.9. Atomic absorption spectrometry

The technique was obtained from Olmedo *et al.*, [38]. The standard solutions to be used for Atomic Absorption Spectrometry (AAS) standard solutions for Manganese (Mn), Nickel (Ni), Aluminium (Al), Zinc (Zn), Chromium (Cr), Iron (Fe), and Lead (Pb) (Titrisol grades) were used to design the calibration curve. They were formed from a stock solution of 1 g/l for each metal ion by successive dilutions with distilled water. Atomic Absorption Spectrophotometer (model 68000, by Shimazu,

Japan) was used. The electrode of the machine was placed inside the beaker containing the prepared sample after the calibration. The absorbance recorded for each metal in a sample was plotted inside the calibration curve to determine the exact concentration of the respective metal.

2.10. Data analysis

Means of various parameters were determined and the statistical difference of means was tested by one-way analysis of variance (ANOVA). Sigmastat v.3.5 was used for the analysis.

3. Results

The collected metal welding fumes were found to be in the respirable size range with a count mean diameter of ${<}1~\mu\text{m}.$

Groups IA-D have not shown any observable changes in their behavior which was similar to the response from the control group. Similarly, groups IIA has shown no observable changes. However, groups IIB & IIC has responded with slight weakness and weakness respectively. Furthermore, group IID has shown slight gasping, prostration, and sluggish movement post-administration. In groups IIIA, IIIB & IIIC they show weakness, prostration, and sluggish response. Group IIID has revealed gasping, apnoea, lack of orientation in movement.

4. Discussion

A study by Reasor and Antoninim [39], revealed that similar results were obtained when the exact quantity of silica was administered at once or spread across five daily separate instillations. Thus, effects from a large bolus of particles seem to be less as long as the dose is not excessively large [39]. For the intratracheal route of administration, there are advantages to its adoption in this study as well. The actual dose of fume administered to each animal is very uniform and can be delivered accurately, without concern for any particles being removed nasally. Experimental designs using intraperitoneal or intratracheal administration are relatively economical. Chronic inhalation experiments, which more closely mimic the type of exposure potentially received by welders, can be very expensive. The dosage used in the current study mimics the derived exposure burden based on assumption using some variables at different rates (2, 4, & 8 h for 3, 5, & 10yrs). This in turn will help in deriving models of dose-response in effects to monitor and regulate activities by relevant authorities. However, this might not depict exactly the response from exposure when compared to inhalation design. Though there are certain advantages of instillation over inhalation exposure, there are some concerns that exist relating its use. First, there is that the introduction of the toxicant is non-physiologic, involving invasive delivery, and the doses are usually larger than what would have been inhaled. Also, there is a difference in the distribution of an instilled material within the respiratory tract and inhaled material. Moreover, the upper respiratory tract that could be an important potential target site during inhalation is bypassed by intratracheal instillation. The medium or vehicle of the test material in which it is suspended or dissolved may influence its distribution in the lungs. Another serious concern is the potential confounding effects of anesthesia, which could affect the initial effect of the instilled material on the lung surface, as well as test material retention and clearance. These limitations could affect the adoption of intratracheal instillation as an acceptable alternative to inhalation [40].

The fumes contained Iron (Fe), Nickel (Ni), Zinc (Zn), Manganese (Mn), Lead (Pb), Chromium (Cr), Cobalt (Co), Cadmium (Cd), Magnesium (Mg), Calcium (Ca), and Potassium (K) with concentrations of 1.894, 0.019, 0.011, 0.013, 0.27, 0.009, 0.18, 0.007, 0.086, 0.006, 0.063 and 0.71 μ g/g respectively as shown in Table 1. The metals are in the sequence of Fe > K > Pb > Co > Cd > Ca > Ni > Mn > Zn > Cr > Al > Cu > Mg with Fe having the highest concentration and Mg with the least.

Table 1

Concentration of heavy metals in welding fumes generated in Kano metropolis.

Metals	Concentration (µg/g)	Limit of Detection (LOD) ($\mu g/g$)		
Iron (Fe ³⁺)	1.8935	0.002		
Nickel (Ni ²⁺)	0.0194	0.002		
Zinc (Zn^{2+})	0.0107	0.001		
Manganese (Mn ²⁺)	0.0132	0.0005		
Lead (Pb ²⁺)	0.2696	0.0005		
Chromium (Cr ³⁺)	0.0089	0.0005		
Aluminium (Al ³⁺)	0.0071	0.001		
Cobalt (Co ²⁺)	0.1803	0.0005		
Copper (Cu ²⁺)	0.0068	0.005		
Cadmium (Cd ²⁺)	0.0854	0.01		
Magnesium (Mg ²⁺)	0.0064	0.001		
Calcium (Ca ²⁺)	0.0628	0.005		
Potassium (K ⁺)	0.7174	0.05		

Beckett [41], stated that electrodes of mild steel (MS) comprised mostly of iron (Fe) with low and differing levels of Mn. Metalworks that utilized stainless steel (SS), aluminum (Al), Ni, and other alloys account for <10 % of all performed metal works. Patti et al., [42], also state that electrodes of stainless steel contain a significant concentration of Cr, in addition to Fe, Mn, and Ni. It has been confirmed in the present study as stainless steel electrode was used, though the concentration of Cr is less than Fe, Mn, and Ni but not much different. Such differences in the quantity of elemental composition could be based on the difference in process of welding employed. Besides, it is revealed that oxides and salts of most other elements might be available in the fumes depending on the metals of welding and processes performed [42].

The main components of the welding fumes are oxides of Fe, Mn, and Si at 41, 18, and 6%, respectively including Cr [43,44]. The majority of welding materials are alloys from mixtures of metals characterized by various steel that may contain Fe, Mn, and Ni. Many animal studies have shown that the presence and combination of different metals is a crucial factor in ascertaining pneumo-toxic responses related to welding fumes exposure. These have described that stain-less steel (SS) welding fumes, which contain significant levels of Ni and Cr, caused more damages to the lungs and they remained in the lungs longer than mild steel (MS) welding fumes, which contain higher Fe [1]. Antonini et al. [45], revealed that some alkali metals such as Potassium (K) maybe be seen in the fluxes that make fumes of manual metal arc welding water-soluble. Ions of Mn have been present in fumes produced from numerous processes, and their proportions were dependent on the welding process and settings [46,47]. Similarly, Cr⁶⁺ was observed to be present and its level may differ significantly depending on the process of welding [48]. Signs of toxicity including clinical symptoms ranged from slight weakness to major weakness (Table 2). Also, slight gasping, prostration, and sluggish movement were eminent in animals exposed to higher doses of fumes. Other symptoms observed at higher doses included gasping, apnoea, and lack of orientation in movement which were signs of

Table 2

Clinical and toxicity symptoms in test animals exposed to lower, medium, and higher doses of metal welding fumes for 12 weeks.

Test animal groups	Clinical signs
IA	No observable signs
IB	No observable signs
IC	No observable signs
ID	No observable signs
IIA	No observable signs
IIB	Slight weakness
IIC	Weakness
IID	Slight gasping; Prostration; Sluggish movement
IIIA	Weakness; Slight gasping
IIIB	Prostration; Slight gasping
IIIC	Weakness; Sluggish movement; Gasping
IIID	Gasping; Apnoea; Lack of orientation in movement
Control	No observable signs

toxicity as observed in Table 2. It can be seen that in Tables 3-5, the concentration of Pb in all groups were higher than the control but were not statistically significant between them and control (p > 0.05). Similarly, Sani & Abdullahi, [49] revealed higher concentrations for Manganese (Mn), Lead (Pb), and Nickel (Ni) in blood. Also, another study found higher Pb and Ni levels in blood and urine samples of welders than the control workers as stated by Iarmarcovai et al. [50], and Botta et al. [51] found out that concentrations of Cd, Co, Ni, and Pb in the blood of welders were higher compared to the control group but not significant. Larmarcovai et al. [50] performed another study to assess the risk associated with welders occupationally by analysis of metals in some biological fluids. However, they found higher concentrations of Pb and Ni in the blood and urine of the two groups of welders compared to controls. Statistically, a difference was found between the welders of group 1 and group 2 in terms of concentration of cobalt in blood and concentrations of Pb and Ni in the urine. Also, the levels of cobalt, lead, and nickel was found to be greater in the blood and urine of welders working in areas without any protection device than those working with smoke extraction systems. Though, the differences that were seen were not all significant statistically. The present study revealed an occupational exposure particularly to lead and nickel. The mean levels of these metals (Pb and Ni) in exposed groups were higher than in controls as reported in several studies on welders [52-56].

In Tables 3–5, there is a statistically significant difference for Cr^{6+} between all the groups (p > 0.05). Results from Ateeq et al. [57] supported the findings of the present study. They found out that there were significantly higher hexavalent chromium levels in all the metal chrome workers when compared to control groups. Similarly, Arshad et al. [58] assessed the concentration of manganese and chromium in blood, urine, and hair of welders and non-welders. From the results, it was clear that manganese and chromium were higher in all the samples analyzed when compared to the international norms set by the world health organization (WHO), (i.e., 20-80 ng/l in blood, 1-8 ng/l in urine, and 300 ng/l in hair for manganese while 20-50 ng/l in blood, 0.24-1.8 ng/l in urine and 100–1000 ng/l in hair respectively for Cr⁶⁺). Iarmarcovai et al. [50] assessed the occupational risk of welders by metals analysis of some biological fluids. There was a higher concentration of chromium in blood and urine in the groups of welders (two groups) as when compared to the control group. Also, there was a statistically significant difference between group 1 and 2 welders for blood cobalt concentration and urine chromium concentration. The concentration of chromium in blood and urine was seen to be higher in welders that are working in areas without protective gear than those working with smoke extraction systems. Nonetheless, the differences observed were not significant statistically. The study revealed an occupational exposure specifically to chromium. The mean levels of chromium in exposed groups were higher than in controls as reported in several studies on welders [52-56]. Sani & Abdullahi, [49], also stated that there were higher concentrations of Chromium (Cr). Also, Jiunn-Liang et al. [59] reported that Cr and Mn levels were significantly greater in welders than they were in administrative workers. A higher concentration of Cr was observed in blood and urine samples of welders more than were seen in control workers [50]. Botta et al. [51] found higher Cr concentrations in the blood of welders compared to in the blood of the control group. Berlinger et al. [60] reported that the water-soluble metal components such as Cr, Cr (VI) in the respirable aerosol fraction were 60%-97% of inhalable aerosols, and 64%-94% of total metal concentration.

Welding fumes of smaller sizes within the nano-size range ($<0.1 \mu m$) induce greater risks for human health. Furthermore, several kinds of research have described that the particle sizes and distribution of welding fumes depend on the combination of other factors which include welding conditions, methods of welding, as well as methods of analysis [43,44,61–64].

Particles generated from welding with rutile coated electrodes were the most toxic because of their high Cr and Ti at 15 and 18.2–20.5%. Moreover, they had 24.5 % of Ni, 1.3 % of Vn, 2.1 % of Mn, and 0.5 % Si.

Table 3

Mean concentration of some heavy metals in the blood of animals exposed to lower doses of welding fumes.

Test animal Groups	Pb (mg/l)	Cr (mg/l)	Fe (mg/l)	Zn (mg/l)	Al (mg/l)	Mn (mg/l)	Ni (mg/l)
IA	0.004 ± 0.001	0.001 ± 0.0002^{a}	$1.67 \pm 0.46^{ m abc}$	0.388 ± 0.14^{a}	0.077 ± 0.025	0.0081 ± 0.0004	0.0096 ± 0.0003
IC	0.008 ± 0.003 0.034 ± 0.005	$0.001 \pm 0.0003^{\circ}$ $0.0015 \pm 0.0011^{\circ}$	$2.18 \pm 1.01^{\circ}$ $2.49 \pm 0.97^{\mathrm{bd}}$	$0.42 \pm 0.11^{ m cm}$ $0.402 \pm 0.17^{ m de}$	0.079 ± 0.031 0.066 ± 0.022	$\begin{array}{c} 0.0071 \pm 0.0003 \\ 0.0089 \pm 0.0006 \end{array}$	$\begin{array}{c} 0.0119 \pm 0.0005 \\ 0.0138 \pm 0.0008 \end{array}$
ID	$\textbf{0.038} \pm \textbf{0.009}$	0.004 ± 0.0023^{d}	2.68 ± 0.73^{ce}	0.35 ± 0.08^{cd}	0.083 ± 0.015	0.0093 ± 0.0005	0.0151 ± 0.0031
Control	0.002 ± 0.0005	0.0003 ± 0.0001^{abcd}	1.34 ± 0.21^{de}	0.34 ± 0.09^{abe}	$\textbf{0.078} \pm \textbf{0.009}$	0.0064 ± 0.0004	0.0043 ± 0.0012
P-value	>0.05	<0.05	<0.05	<0.05	>0.05	>0.05	>0.05
LOD	0.0005	0.0005	0.002	0.001	0.001	0.0005	0.002

p < 0.05: there is significant difference.

Table 4

Mean concentration of some heavy metals in the blood of animals exposed to medium doses of welding fumes.

Test animal Groups	Pb (mg/l)	Cr (mg/l)	Fe (mg/l)	Zn (mg/l)	Al (mg/l)	Mn (mg/l)	Ni (mg/l)
IIA IIB IIC IID Control	$\begin{array}{c} 0.006 \pm 0.001 \\ 0.007 \pm 0.0015 \\ 0.0105 \pm 0.003 \\ 0.036 \pm 0.008 \\ 0.002 \pm 0.0005 \end{array}$	$\begin{array}{c} 0.002\pm 0.0007\\ 0.0023\pm 0.0008^{a}\\ 0.0015\pm 0.0002^{b}\\ 0.0005\pm 0.0001^{c}\\ 0.0003\pm 0.0001^{abc} \end{array}$	$\begin{array}{c} 1.83 \pm 0.67^{a} \\ 2.49 \pm 0.41^{b} \\ 2.58 \pm 0.53^{ac} \\ 2.98 \pm 0.31^{d} \\ 1.34 \pm 0.21^{bcd} \end{array}$	0.36 ± 0.04 0.402 ± 0.02 0.35 ± 0.05 0.38 ± 0.02 0.34 ± 0.09	$\begin{array}{c} 0.065 \pm 0.003 \\ 0.068 \pm 0.002 \\ 0.078 \pm 0.003 \\ 0.079 \pm 0.002 \\ 0.078 \pm 0.009 \end{array}$	$\begin{array}{c} 0.0088 \pm 0.0002^{a} \\ 0.0092 \pm 0.0002 \\ 0.0097 \pm 0.0004 \\ 0.0122 \pm 0.0011^{ab} \\ 0.0064 + 0.0004^{b} \end{array}$	$\begin{array}{c} 0.0102 \pm 0.0009 \\ 0.0133 \pm 0.0022 \\ 0.0149 \pm 0.0032 \\ 0.016 \pm 0.0021 \\ 0.0043 \pm 0.0012 \end{array}$
P-value LOD	>0.002 ± 0.0003 >0.05 0.0005	<0.005 0.0005	<pre></pre>	>0.05 0.001	>0.078 ± 0.009 >0.05 0.001	<0.0004 ± 0.0004 <0.05 0.0005	>0.0043 ± 0.0012 >0.05 0.002

p < 0.05: there is significant difference.

Table 5

Mean concentration of some heavy metals in the blood of animals exposed to higher doses of welding fumes.

Test animal Groups	Pb (mg/l)	Cr (mg/l)	Fe (mg/l)	Zn (mg/l)	Al (mg/l)	Mn (mg/l)	Ni (mg/l)
IIIA IIIB IIIC IIID Control P-value	$\begin{array}{c} 0.0195 \pm 0.003 \\ 0.007 \pm 0.001 \\ 0.0105 \pm 0.002 \\ 0.0165 \pm 0.003 \\ 0.002 \pm 0.0005 \\ \textbf{>}0.05 \end{array}$	$\begin{array}{c} 0.0025 \pm 0.0011 \\ 0.0015 \pm 0.0007 \\ 0.0027 \pm 0.001^a \\ 0.0035 \pm 0.0014^b \\ 0.0003 \pm 0.0001^{ab} \\ < 0.05 \end{array}$	$\begin{array}{c} 2.27 \pm 0.78 \\ 2.68 \pm 0.62^a \\ 3.13 \pm 0.58^b \\ 3.42 \pm 0.22^c \\ 1.34 \pm 0.21^{abc} \\ < 0.05 \end{array}$	$\begin{array}{c} 0.323 \pm 0.003 \\ 0.37 \pm 0.002 \\ 0.414 \pm 0.009 \\ 0.441 \pm 0.012 \\ 0.34 \pm 0.09 \\ \color{red} > 0.05 \end{array}$	$\begin{array}{c} 0.053 \pm 0.003 \\ 0.078 \pm 0.002 \\ 0.079 \pm 0.004 \\ 0.089 \pm 0.0033 \\ 0.078 \pm 0.009 \\ \textbf{>}0.05 \end{array}$	$\begin{array}{c} 0.0076 \pm 0.0001^{a} \\ 0.0097 \pm 0.0002^{b} \\ 0.0109 \pm 0.0007^{c} \\ 0.0141 \pm 0.0031^{d} \\ 0.0064 \pm 0.0004^{abcd} \\ < 0.05 \end{array}$	$\begin{array}{c} 0.0113 \pm 0.0001^{ae} \\ 0.0144 \pm 0.0001^{b} \\ 0.0172 \pm 0.0001^{c} \\ 0.0220 \pm 0.0001^{de} \\ 0.0043 \pm 0.0012^{abcd} \\ < 0.05 \end{array}$
LOD	0.0005	0.0005	0.002	0.001	0.001	0.0005	0.002

p < 0.05: there is significant difference.

However, some particles formed by welding with electrodes having rutile-cellulose coating among all samples studied were the least toxic because they do not contain Cr and Ti was 40.1–46.6 times less than in particles with rutile coating [65]. Commonly used electrodes with different coatings were used and maximum pollution occurred with PM10 fraction during arc welding operations. It has also demonstrated excessive concentrations of PM10 particles at distances 0–3 m and 4–5 m from the emission source [66].

From Tables 3–5 there is a significant difference between the groups (p < 0.05) in terms of Fe³⁺. Lu et al. [67] reported that meanwhile there was no statistically significant difference for serum transferrin receptor levels, serum manganese, iron, ferritin, and transferrin content in welders were found to be greater than the control group as similarly reported in this present study. A positive relationship was found to exist between serum iron and ferritin levels and the working period. Based on their results, the origin of exposure to metal fumes changes the amount of serum iron, manganese, and iron metabolism-related proteins [67]. In the present study, Iron been one of the metals with the highest composition in welding fumes can alter the blood iron levels thereby supporting the study by Lu et al. [67]. Berlinger et al. [60] reported that the water-soluble metal components such as Fe in the respirable aerosol fraction were 60%–97% of inhalable aerosols, and 64%–94% of total metal concentration.

From Table 3, there is a significant difference statistically between the groups (p < 0.05). However, in Table 4 and 5, there is no significant difference statistically between the groups (p > 0.05) in terms of Zn. In a study by Abdulrahman et al. [68], they determined the levels of heavy metals that are related to health hazards particularly Zn in samples of nails among car workshop workers. The results of this study showed that welding workers revealed the highest levels of heavy metals in their nail samples while in car mechanics it was the lowest levels. They also reported that the age of the workers might have influenced the heavy metals concentration positively especially for Zn and Pb, 20.9, 6.8 μ g/g, respectively.

In another study on 28 short-term exposure of 21 chronic exposed and 33 control individuals whereby saliva samples were collected for analysis of manganese, copper, zinc, cadmium, and lead levels. The concentration of Zn in Welders' salivary was found to be lower while manganese and copper levels were higher compared to controls. The levels of cadmium and lead in the saliva did not vary. The period of exposure correlated with saliva contents and saliva levels of manganese might be a marker for occupational welding fume exposure and working period [69].

From Tables 3–5, there is no significant difference statistically between the groups (p > 0.05). However, in another study by Iarmarcovai et al. [50], there was a statistically significant difference between groups 1 and 2 of welders for the concentration of Al. Exposure to such pollutants for the long term is enough to modify the physiological process in the body of workers [70]. Hasan et al. [71] mentioned in their study that the occupational exposure is a much complex phenomenon due to many reasons that contribute to the absorption and depletion of metals; such factors include illness, poor diet as well as person susceptibility [71]. It might also be due to their specificity in terms of metabolic processes which may cause different levels of intake and accumulation in workers' body tissues [72].

From Table 3, there was statistically no significant difference between the groups (p > 0.05). However, in Tables 4 and 5 there is a statistically significant difference between the groups (p < 0.05) in terms of Mn. Similarly, Jiunn-Liang et al. [59] reported a significantly higher concentration of Mn in welders than they were observed in administrative workers. Also, Sani & Abdullahi, [49], revealed higher blood and urine concentrations for Manganese (Mn). These metals were significantly higher in welders than when compared to the control as was stated by many kinds of research [52–56]. Also, Mn levels in the blood of welders (16.6 µg/L) and administrative workers (14.0 µg/L) in the study by Jiunn-Liang et al. [59] were greater than those found in factory workers (5.5 µg/L), a control group in Taiwan (5.4 µg/L) [73], and South Korean adults (10.8 µg/L) [74].

From Tables 3 and 4, there is no significant difference between the groups (p > 0.05). However, in Table 5, there is a significant difference between the groups (p < 0.05) in terms of Ni. Similarly, Sani & Abdullahi, [49] stated that there was a higher concentration of Ni in the blood and urine of metal workers than in control. Botta et al. [51] reported higher concentrations of Ni in the blood of welders when compared to that of the control group. Iarmarcovai et al. [50] also reported higher levels of Ni in blood and urine in the two groups of welders compared to controls. Statistical difference was observed between the welders of group 1 and group 2 for urine concentrations of Ni. Also, blood and urinary concentrations of Ni was found to be higher in the welders working in areas without any protection device than those working with smoke extraction systems. Nevertheless, the differences were not statistically significant. The mean levels of these metals in exposed workers groups were higher than in controls as reported in several studies on welders [52-56]. Abdulrahman et al. [69] measure the concentration of Ni in nail samples of car workshop workers. They revealed that welding workers recorded the highest concentration levels in their nail samples meanwhile car mechanics revealed the lowest levels. Ni levels were found to be the highest compared to other heavy metals.

5. Conclusion

The metal fumes analyzed contained heavy metals which are in the sequence of Fe > K > Pb > Co > Cd > Ca > Ni > Mn > Zn > Cr > Al > Cu > Mg. Changes were noticed in terms of behavioral activities of the test animals compared to the control indicating probable toxicity. The values of Mn, Ni, Pb, Cr, and Fe in the exposed animals are higher than the control and increased relatively across the treatment groups. However, the values of Al and Zn were not significantly different from the control. These showed that exposure to welding fumes having contained a significant amount of heavy metals has caused noticeable toxicity symptoms and a simultaneous elevation in corresponding levels of metals in the blood.

Authorship contributions

Category 1

Conception and design of study: I.L. Abdullahi, A. Sani. Acquisition of data: A. Sani, I.L. Abdullahi.

Analysis and/or interpretation of data: A. Sani, I.L. Abdullahi.

Category 2

Drafting the manuscript: A. Sani, I.L. Abdullahi.

Revising the manuscript critically for important intellectual content: I.L. Abdullahi, A. Sani.

Category 3

Approval of the version of the manuscript to be published (the names of all authors must be listed): I.L. Abdullahi, A. Sani.

Declaration of Competing Interest

The authors report no declarations of interest.

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