Original article:

Serum heavy metal levels in teenagers currently or formerly employed as gas station attendants

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Abstract

Background: Reports are available indicating that leaded gasoline is still being made available in some parts of the world. Moreover it is also known that the crude from which petroleum products are obtained is rich in heavy metals. This means heavy metal toxicity from petrol contact is a possibility. The aim of this study is to assess selected heavy metal levels in teenagers who are currently and formerly employed as GSA. ***Materials and Methods:*** The study population consisted of teenagers divided into 3 groups. GROUP I was made up of 30 male subjects that are currently employed as GSA with less than 6 months work history; GROUP II composed of 34 male teenagers with work history in gas station not less than 20 months; GROUP III consisted of 22 male teenagers who were former attendants; the control group consisted of 35 participants, with no history of occupational exposure to fuel, or heavy metals. Information was obtained through questionnaire on demography, life-style and clinical symptoms of gasoline exposure. Serum heavy metal levels were estimated using atomic absorption spectrometry. Student's t test and analysis of variance were employed to determine the level of significant difference. $P < 0.05$ was considered significant. ***Results and Discussion***: The serum levels of lead, arsenic, nickel, cadmium, and aluminium of GSA in GROUPS I and II were significantly higher compared with control. On the other hand, there were significant increases in levels of lead and cadmium in GROUP III compared with control. Additionally, majority of test subjects in GROUPS I and II identified headache and fatigue as symptoms associated with workplace. Teenage GSA are prone to heavy metal toxicity and discontinuing contact with gas station environment did not resolve this occupational hazard. ***Conclusion***: Gonadotoxic effect of some of these elements on these categories of subjects who are within reproductive stage is possible. Therefore appropriate measure should be taken to address it.

Keywords: cadmium; lead; arsenic; aluminium; gas station attendant.

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Introduction

Many of the studies that have been conducted on gas station attendants (GSA) have been directed at estimating the levels of gasoline components (usually benzene, toluene, xylene) in the blood samples of these subjects as well as in the filling station environment and correlating the results obtained to the degree of toxicity induced in exposed attendants[1,2]. While it is true that gasoline is made up of different types of hydrocarbon compounds, the crude oil from which it is obtained contains a number of heavy metals[3,4.]Moreover many additive metals are incorporated into different fuel types to enhance their performance[5-7.] Although the incorporation of tetraethyl lead to gasoline has been discouraged, few reports still maintain that there seems to be a high level of lead in gasoline made available in some parts of the world[8.] Moreover, the crude is known to contain metals like V, Ni, Cu, Cd and Pb[3.] Because of the possibility of fuel exposure to these attendants especially by different routes (nasal-from vapour; dermal-from spillage), it is expedient that the serum levels of Pb, Cd, Si, As, Al and Cd in petrol attendants be determined.

Several of the components of gasoline have significantly hazardous carcinogenic properties. Human contact to gasoline occurs from its significant release as vapours, and this has been identified as major source of exposure in the large number of fuel refilling stations and adjoining residential areas[9.] Both epidemiological and experimental studies have confirmed the carcinogenic and mutagenic properties of petrol and diesel fumes[10.] Many carcinogens require that they be exposed to an animal for extended period

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of time before the carcinogenic process is initiated, an example of this is aflatoxin B1. Yet the hepatoxic effect of aflatoxin with short-time duration of exposure especially when large quantities are administered to an experimental animal has been documented also[11.] Most of the studies that have been carried out to evaluate the impact of occupational exposure to gasoline involved study populations who have been exposed for extended periods of time usually in years, with several of these studies showing significant alteration in many metabolic processes.

The aim of the present study is to investigate the impact of short-time exposure to petroleum products on heavy metal metabolism in petrol attendants with less than 6 months of exposure period and compare it longer period of exposure (>20 months). In addition, by including former GSA, it will be possible to identify whether heavy metal toxicity that occurs from contact with petrol is resolved from cessation of exposure.

Materials and methods

Selection of participants

The study was conducted in subjects that were recruited within Ibadan metropolis. The study population consisted of teenagers divided into 3 groups. GROUP I was made up of 30 male subjects that are currently employed as GSA with less than 6 months work history; GROUP II composed of 34 male teenagers with work history in gas station not less than 20 months; and GROUP III consisted of 22 male teenagers who were former attendants. The following demographic information was obtained from each subject: age, length of employment/ duration of exposure to petroleum products, number and duration of shift per week, any particular symptoms associated with workplace. Other information obtained was on lifestyle (smoking, drinking, coffee). All the gas station attendants recruited for the study were essentially involved in pumping petrol and diesel into vehicles. They were asked about the type of safety measure employed e.g. facemasks or hand gloves. The control group also filled similar questionnaire and provided all necessary information. The control group consisted of 35 subjects from the general population with no history of occupational exposure to fuel, or heavy metals. The test and control subjects were age-matched. Informed consent was obtained from each individual prior to the commencement of the study. The study was carried out in accordance with revised Helsinki declaration. The study was approved by the local ethics review board.

Sample Collection and heavy metal analysis:

From each participant five millimeters of blood was collected into a tube without anticoagulant. For GROUPS I and II, the blood was collected at the end of at least 8-h shift, whereas for GROUP III and control, blood was collected around the same time as GROUPS I AND II to allow for uniformity. In each case blood sample was allowed to clot and then centrifuged at 2500 g for 10 minutes to obtain serum for biochemical analysis. The serum obtained was used to estimate the concentrations of lead, cadmium, aluminium, silicon, nickel, and arsenic. The method of atomic absorption spectrometry was employed. Buck Scientific 205 Atomic Absorption obtained from Buck Scientific located in East Norwalk, Connecticut (USA) was used for these analyses.

Statistical analysis: Statistical Package for Social Sciences (version 15) was used for the analysis of data to obtain mean ± SD (standard deviation). The level of significant difference between exposed teenage subjects and age matched control was assessed by Student's t test and analysis of variance. $P \leq 0.05$ was considered significant.

Results

The average number and duration (range) of shift per week for GROUPS I- III were 5 and 7-9 hours respectively. Majority of GSA in GROUP I (25 participants) and GROUP II (29 participants) identified headache and fatigue as symptoms associated with workplace, although they are of the opinion that these symptoms may be linked to standing on their feet for prolonged period of time in the course of their daily duty, rather than from environmental contamination of the filling stations occurring from volatile nature of gasoline. 7 of GSA in GROUP III indicated that they still suffered (infrequently) from these symptoms. Not less than 35 (participants) of all test subjects revealed that when they first took up employment at fuel filling station, sneezing or nausea were pronounced at the commencement of work each day, but that the two symptoms disappeared not later than 2 weeks after taking up appointment at the stations, while 9 GSA of the test population reported that they presented with cough within the same period. None of the subjects (test and control) was a habitual smoker, or alcohol and coffee consumer; none had taken alcohol or smoked in the past three years prior to the time that they were recruited for the study. While many of the test subjects revealed that the safety measures (face mask glove, overall) were not always available in their workplace, those who could have accessed them did not see the need to use these measures because they were of the opinion that although gasoline is volatile, it was probably not dangerous at low dose exposure.

The results of serum analyses of control, GROUPS I and GROUP II are shown in **Table 1**. Serum levels of nickel, aluminium, arsenic, lead and cadmium showed significant increases in both GROUPS I and II compared with control; silicon was additionally increased in GROUP II subjects. GSA formerly employed in gas station had significant increases in many of the estimated elements but others were not significantly different compared with control values as shown in Table 2. The levels of Ni, Pb, Cd, and Al were comparable in both GROUPS I and III as there were no significant difference between the two groups but As and Si were significantly lower in GROUP III compared with GROUP I. On the other hand, As and Si of GROUP III were not significantly different from control as revealed in Table 2.

Table 1: Serum levels of heavy metals of control and gas station attendants with different duration of exposure.

	Ni (µg/L)	As (ng/dL)	Al (ng/L)	Si (ng/L)	Pb (µg/L)	Cd (mg/dL)
Control	0.62±0.21	0.81±0.30	12.00±4.23	160.19±30.26	0.15±0.03	0.20±0.04
GROUP I	0.87±0.12*	0.99±0.36*	17.00±8.02*	169.00±26.64	0. 32±0.07*	0.29±0.04*
GROUP II	0.94±0.20*	0.85±0.19	18.65±3.63*	161.29±10.21	0. 41±0.10*	0.44±0.104*
F value	6.103	10.252	7.145	2.326	4.091	23.915
P value	0.009‡	0.014‡	0.011‡	0.122	0.018‡	0.007‡

Results are expressed as mean ± standard deviation. *p value significant when compared with control. ‡p value significant using analysis of variance.

Table 2: Serum levels of heavy metals of control, current and former gas station attendants.

	Ni (µg/L)	As (ng/dL)	Al (ng/L)	Si (ng/L)	Pb (µg/L)	Cd (mg/dL)
Control	0.62±0.21	0.81±0.30	12.00±4.23	160.19±30.26	0.15±0.03	0.20±0.04
GROUP I	0.87±0.12*	0.99±0.36*	17.00±8.02*	169.00±26.64	0. 32±0.07*	0.29±0.04*
GROUP III	0.80±0.41*	0.82±0.32	15.40±8.02*	159.52±38.09	0.30±0.11*	0.29±0.17*
F value	11.630	23.041	80.113	9.287	53.610	40.909
P value	0.010¶	0.039¶	0.023¶	0.274	0.017¶	0.031¶

Results are expressed as mean ± standard deviation. *p value significant when compared with control. ¶p value significant using analysis of variance.

Discussion and conclusion

When the level of genetic damage was evaluated through micronucleus test (MNT) in buccal epithelial cells (BEC) and chromosomal aberrations (CA) assay in peripheral blood lymphocytes of gas station attendants (GSA) by Rekhadevi et al.[12.] The results of the study showed that contact with BTX (benzene, toluene, xylene) in petrol vapors induced a statistically significant increase in the frequency of micronuclei (MN) and CA in the exposed subjects compared with control. This is not surprising, fuel (diesel and petrol) constitutes a complex mixture of volatile flammable liquid hydrocarbon; the very hazardous of the hydrocarbon constituents are benzene, toluene, and xylene; benzene especially is considered the most toxic of them because of its carcinogenic potency. Exposure to these compounds may have a negative effect on the health of the exposed subjects. The kind of presentation reported by Rekhadevi et al.[12.] while it definitely suggests the harmful effects of gasoline exposure, it does not prove that those effects were exclusive to BTX.

The results of the present study raise the possibility of the involvement of non-essential metals in many of the pathologic processes that have been associated with gasoline exposure in GSA. Many heavy metals are found in petroleum products either originating from the crude from which they are obtained or those introduced in the course of production to enhance their efficiency. Take for instance the study of Onunkwor et al.[8], in order for them to determine the ameliorative effects of vitamin C in chronic lead poisoning, petrol station attendants and auto-mechanics in Abeokuta, who have been shown to be occupationally exposed to lead and university students (serving as control), were supplemented daily with 500 mg vitamin C acid for 2 weeks. Prior and subsequent to vitamin C regimen, blood and urine samples were collected from the subjects and analysed for lead and biochemical effects associated with lead toxicity. After 2 weeks of vitamin C supplementation, there was significant decrease in blood lead in the occupationally exposed subjects. The reduction in blood lead was as much as 57% in male petrol station attendants, 50% in female petrol station attendants and 44% in the auto-mechanics. In addition, lead urine content was elevated significantly in the occupationally exposed subjects. The biochemical effects usually linked with lead toxicity responded positively to the ascorbic acid supplementation. Plasma and urine aminolevulinic acid (ALA) were reduced significantly by as much as 55% and 57% respectively. The metabolism of vital element- calcium was also modulated; Vitamin C supplementation equally resulted in significant increase in plasma calcium level in test subjects compared with control. It also reversed decreased levels of reduced glutathione (GSH) and hemoglobin observed in the occupationally exposed subjects, although glutathione-S-transferase (GST) and catalase (CAT) activities were not affected. More importantly, Onunkwor et al.[8] also revealed a significantly high level of lead in petrol station attendants than control subjects. Their study gave credence to the fact that the gasoline made available in some parts of the world is still heavily laden with lead.

While it is generally assumed that the presence of lead in gasoline indicate its incorporation to fuel as additive, the high level of other metals identified in these attendants suggest that its crude oil origin may also be a huge contributing source. Although it should be emphasized that lead is not the only additive metal in petroleum products; several other metals that have been identified to be added to petrol, diesel, and engine oil in different concentrations, include Al, Ni, and Pb evaluated in this study as well as Zn, P, Ca, S, Mg, Mo, Ca, S, P, Zn, Nb, W, Ti and Na that were not determined[13]. The significant increase therefore in the serum level of aluminium, nickel, cadmium, arsenic and lead in current GSA compared with control may be linked to direct contact with these products.

The role that duration of exposure plays in degree of toxicity can be observed from the results of study; when GROUP I was compared with GROUP II, there were significant increases in levels of many elements in serum of GROUP II (GSA with at least 20 months exposure) compared with GROUP I (GSA with less than 6 months exposure). These observations should be of concern because the heavy metals that were significantly elevated in different groups of GSA are well known for their hazardous effects on different organs of the body even with minimal exposure level. More importantly results of the study also revealed many of the metals remained elevated in former GSA which gives credence to what is well known that even when exposure ceases for many elements, the body burden remains high because they are accumulated in tissues. This invariably affects their biological half-lives. The biological half-life of a metal is an indication of its retention and it varies from metal to metal, as well as tissue to tissue. For instance, the biological half-lives of cadmium and lead in kidney and lead respectively are between 20–30 years but with arsenic, the biological half-life is a few hours to days. This may explain why arsenic is significantly lower in former GSA compared with GSA currently in employment. Of particular interest are those metals which require more than one half-life for their retention to be fully described. The half-life of lead in blood is only a few weeks, whereas it has a much longer half-life in bone. In most cases this is true of acute exposure; continued metal exposure clearly complicates retention kinetics. Yet this may explain the elevated levels of Pb in former GSA still recorded after cessation of contact. While blood, urine, and hair are considered appropriate tissues for quantifying metal toxicity being the most accessible samples, data obtained from single measurements may reflect recent exposure or long-term or past exposure, depending on retention time in the particular tissue. Blood concentrations usually, but not always, reflect more recent exposures and correlate with acute adverse effects. This may be a shortcoming of this study. The level of arsenic is significantly lower in former attendants, even though arsenic is well absorbed from the gastrointestinal tract, distributed throughout the body and then excreted primarily in urine. Skin also has been identified as a potential route of exposure to arsenic, and systemic toxicity has been reported in persons having dermal contact with solutions of inorganic arsenic. The significantly low arsenic level can be ascribed to excretion through the urine. Excretion of absorbed arsenic is principally through the urine. The whole-body biological half-life of ingested arsenic is about 10 hours, and over 72 hours as much as between 50 and 80% is excreted. Arsenic has a preference for skin and is eliminated from the body desquamation of skin and in sweat, particularly during periods of profuse sweating. Since it concentrates in formation of fingernails and hair, the significant low level may not necessarily reflect body-burden of arsenic. Tunsaringkarn et al.[14] revealed that the most common symptoms associated with gasoline exposure were headache, fatigue and throat irritation. However, the present study identified not only headache and fatigue as symptoms of occupational exposure to gasoline, but cough, nausea, and sneezing as well. These findings are in consonant with results of a previous study, which revealed that low concentrations of benzene caused drowsiness, dizziness, and headaches[14,15.] The study of Tunsaringkarn et al.[14] revealed that benzene and toluene exposures were significantly associated with fatigue. However, many of the heavy metals found to the significantly higher in studied population compared with control have been reported to affect the hemopoetic system, inducing low level of hemoglobin and by implication anemia. Studies have revealed that there was an inverse correlation between degree of fatigue and hemoglobin level. Different immunological abnormalities have been reported in patients with fatigue. For example, it has been noted that immune aberrations elevates the decay of the stress protein mRNAs via proteins 2-5A synthetase (2-5A) and protein kinase RNA (PKR) activity in those exposed to benzene. While sufficient evidence exists to suggest that chronic fatigue syndrome has the involvement of abnormal immune system but in the present study the immune markers were not assessed. Therefore, a correlation between immune markers, serum heavy metal levels and degree of fatigue could not be determined. This study demonstrates that the attendants are at risk of exposure to heavy metals even when exposure period does not exceed 6 months. While the high serum BTX level in attendants not using protective gear observed from past study is understandable since BTX are volatile, the results of high levels of heavy metals that are non-volatile could have arisen from exposure through the other routes. Splashing of petroleum products commonly occurs from sale of gasoline into jerry-cans during its period of scarcity. This may be one of the hazards linked with incessant petroleum scarcity in Nigeria.

Conflict of interest: None declared

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