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A Baseline Study for Cadmium Concentrations in Blood of Goats in Some Communities of Bade, Northern Yobe, Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. Author MMG designed and carried out the study, performed the statistical analysis and wrote the first draft of the manuscript. Authors JK and MMS supervised and managed the analyses of the study. Author HIA assisted in the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aim: This study was carried out to evaluate levels of Cadmium (Cd) in goats as sentinels for Cd pollution in some farming communities in Bade Northern Yobe, Nigeria.
Study Design: The study was a cross sectional study.
Methodology: A total of 356 blood samples were collected from semi-intensively managed goats from 5 randomly selected wards in the study area. A total of 55 goats were sampled from Dagona, 72 from Katuzu, 78 from each of Lawan Musa and Sabon Gari and 73 from Usur/Dawayo wards. Sexes of the animals were noted during sampling. Concentration of blood cadmium was assayed

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using Graphite Furnace Atomic Absorption Spectrophotometry (GFAAS). The samples were digested using NH_2:H_2O_2 wet digestion technique. Mean (±SEM) blood Cd concentrations of the sampled goats were 7.677±0.53, 3.835±0.47, 3.996±0.45, 2.810±0.45, and 3.314±0.46 µg/L for Dagona, Katuzu, Lawan Musa, Sabon Gari and Usur/Dawayo wards respectively. Goats from Dagona ward had significantly (P<0.05) higher mean blood Cd concentrations compared to other locations but there was no statistically significant difference in mean blood Cd levels between males and females.

Conclusion: The detection of Cd in the blood of most of the animals (99.4%) is suggestive of how widely distributed Cd is in the area and presence of Cd in blood of 349 (98.6%) goats at concentrations above the recommended normal levels of 0.185 µg/L by the Agency for Toxic Substances and Disease Registry (ATSDR) is worrisome. There is need to identify possible Cd emission sources in the study area in order to control contamination of the environment and hence minimize humans' and animals' exposure to the nephrotoxic metal.

Keywords: Cadmium; goats; Yobe state; Northeastern Nigeria; pollution; graphite furnace atomic absorption spectrophotometry.

1. INTRODUCTION

There is a growing concern over an increasing incidence [1] of chronic kidney disease of unknown etiology (CKDu) in some communities along the coast of river Yobe. Hospital records from the area have indicated increased incidence of CKDu amongst people most especially those between the ages of 20 to 40 years particularly in the last 15 to 20 years [1]. The actual cause of the disease is not known yet and its epidemiology remains poorly understood most probably due to poor health facilities and lack of qualitative health records [2]. The area is located within Komadugu Yobe Basin (KYB) and its, significantly agrarian, population heavily depend on River Yobe as source of livelihoods [3]. Fishery, livestock, irrigation and rain-fed farming practices are their major agri-based activities throughout the year [4]. Previously, there were reports expressing concern over deteriorating quality of the basin's environment most especially due to dumping of industrial and municipal wastes at the upstream Kano river and from the large scale irrigation project at the Hadejia valley [5,6,7].

Wastes from anthropogenic activities have been reported as major sources of toxic heavy metals in the environmental [8]. Nephrotoxic metals such as Cadmium (Cd), Lead (Pb), Arsenic (As) and Mercury (Hg) are some of the metals associated with anthropogenic activities [9,10,11]. They have been reported to cause varying degrees of nephrotoxicity following exposure via food chain and environmental media [12]. Previous researches on different food substances and environmental media from the study area have reported violative concentrations of Cd and Pb. National Agency for Food, Drugs Administration and Control (NAFDAC) [13] have reported concentrations of Cd and Pb in samples of drinking water from the area ranging from 0.01 to 0.10 ppm and Not Detected to 0.30 ppm for Cd and Pb respectively. Similarly, Oyekanmi et al. [14] have reported high concentrations of As, Cd, Pb, Nickel (Ni) and Chromium (Cr) from water sources used for irrigation in Jawa, a village located in the study area. Oyekanmi et al. [14] and Waziri and Lawan, [15] have also reported high concentrations of Cd and Pb in vegetables raised from the study area. Analysis of soil and dust storm from the area also showed high concentrations of Cd and Pb [14,16]. Gashua et al. [17], have also reported elevated levels of Cd and Pb in freshwater catfish from river Yobe. These findings could be good indicators of heavy metals pollution status of the environment and, possibly, its associated risks. The focus of the current research was placed on single metal due to financial constraints. However, cadmium was priorities due to the role it plays in the development of chronic kidney diseases even at low level environmental exposure [18].

Cadmium is a heavy metal with high toxic potentials, even at very low exposure levels, resulting in acute and chronic effects on health and environment [19]. It is relatively a rare element and its most common natural sources in the environment are volcanoes and weathering [20]. Manmade activities play vital roles in the emissions and spread of Cd, for instance, total global anthropogenic air emissions in 1995 was estimated at 3000 tons [19] and in the year 2000, about 19,700 tons of Cd was extracted from the earth's crust by man [21]. Cd can also be

transported over a long distance by air as very fine particles and deposited in areas far away from the emitting source. Cd is naturally not degradable and as such has accumulative effects in the environment [19]. Cadmium and cadmium compounds are more water soluble compared to other metals, and are therefore, more mobile in environmental medium, more bioavailable and tend to bioaccumulate [19]. Animals and non-smoking humans can get exposed to Cd via inhalation of polluted air, consumption of contaminated feedstuffs or food and drinking of polluted water [22,23,24,25,26]. Ruminants can additionally get exposed to Cd through consumption of contaminated soil either accidentally while grazing or intentionally during mineral deficiencies [26]. Dogs, cats and other carnivorous animals can get exposed to violative levels of Cd by ingestion of contaminated meat such as liver and kidneys especially from older animals; these organs are known to contain high concentrations of Cd [27]. Aquatic organisms readily absorb free ionic Cd directly from their water environment [28].

Exposure to Cd can lead to acute or chronic toxicities in humans. Acute toxicity usually occurs following occupational exposure to Cd^{2+} dust or fumes. Inhalation of Cd^{2+} could lead to onset of diffuse pneumonitis associated with severe pulmonary oedema, mucle weakness, chemical pneumonitis which is usually followed by respiratory failure and death [29,30]. Acute toxicity associated with ingestion of Cd can lead to death due to extensive liver and kidney damages [29,31,32]. Chronic exposure to Cd has been reported with varying consequences depending on route of exposure. Oral exposure may lead to renal damage [33,34,35] and severe gastrointestinal irritation with nausea and vomiting especially following exposure to high amount of Cd^{2+} [36]. Pulmonary symptoms are linked to inhalation exposure. Exposure to Cd has also been associated with bone toxicities [31,37,38,39,40], reproductive toxicities [41,42,43,44,45], cardiovascular toxicities [46] and carcinogenic effects [47]. Clinical cases resulting from chronic exposure to Cd are most often misdiagnosed probably due to the fact that it accumulates in the body over a long period of exposure and as such are neglected as primary cause [48].

The aim of this study is to assess the levels of Cd in goats as sentinels for human exposure. Goats are among the most abundant and widely distributed species of livestock grazing freely in the study area [49,50,51], as such they could be good indicators of general environmental pollution profile of the study area [52,53,54]. Blood from animals and humans have been used throughout the world to monitor exposure to heavy metals from the environment [54,55,56,57].

2. MATERIALS AND METHODS

2.1 Study Area

Bade Local Government Area, Yobe State is located between longitude 12º52´N and latitude 10º58´E, situated in Yobe North senatorial district with its headquarters in Gashua. It has an area of about 772 KM^2 (298 sq. Miles) and a population of 139,804 [58]. The LGA has ten political wards, six of which are located within Gashua, headquarters of the LGA *viz* Lawan Musa, Sarkin Hausawa, Lawan Fannami, Sabon Gari, Zango and Katuzu, whereas the other four wards are made up of districts or clusters of villages and settlements around Gashua, they include Usur-Dawayo, Dagona, Sugum-Tagali and Gwiokura.

2.2 Study Population

Goats raised in Bade local government area are the subjects selected for this study. Goats which are more than six months of age or have spent at least three months in the study area were selected for sampling. Blood sample was collected from both sexes.

Sampling Procedure: Multi-stage sampling technique was used as follows: -

Stage 1: 50% of the 10 wards were selected using random numbers generated by Microsoft excel software program. It comprises of 3 wards from Gashua namely Katuzu, Lawan Musa and Sabon Gari, and 2 from the rural areas namely Dagona and Usur/Dawayo.

Stage 2: 25% of households which keep goats in the selected Gashua wards were targeted for sampling whereas, all accessible villages under the 2 rural wards were targeted for sampling. Under Dagona, 6 villages were identified namely Bizi, Dagona, Dala, Garin Manda, Mainiya and Misilli whereas in Usur/Dawayo ward have 9 villages were sampled namely Aiso, Alagarno, Azam Kura, Azbak, Bida, Dawayo, Jigawa, Paga and Usur. Sampling from the households was based on consent and from houses that have more than two goats. A minimum of two and

maximum of five (2-5) animals were sampled per household and the goats were selected based on simple random method. Samples collected from each ward were pooled and mean concentrations were used as representative values for each ward.

2.3 Sample Collection

A total of 356 blood samples were collected from the study area; 78 (21.9%) samples each from Lawan Musa and Sabon Gari wards, 72 (20.2%) from Katuzu ward, 55 (15.4%) from Dagona ward and 73 (20.5%) from Usur/Dawayo ward. Three milliliter of blood was taken from each animal via the jugular vein using 5ml disposable syringe and 21G″ needle. Prior to blood collection, point of puncture on the skin was sterilized using cotton wool soaked in commercially prepared methylated spirit. Each blood sample was transferred to a sterile EDTA sample bottle and the tube inverted several times to mix blood with the EDTA. All samples were labeled accordingly and immediately preserved in an ice packed container at about 4°C before transportation to Chemistry Laboratory at the Department of Basic Sciences, Umar Suleiman College of Education Gashua for digestion.

2.4 Sample Digestion

Wet digestion of blood was carried out according to modified procedures described by Shaw et al. [59], Hogstrand et al. [60], and U.S. EPA [61]. Exactly 2.5 bml of whole blood was aspirated from the original sample and transferred unto a 250 ml glass beaker containing 10ml trace metal grade concentrated HNO_3. This was then placed on a hot plate at 100°C until a clear mixture was obtained; it was then allowed to cool at room temperature. Then 1.5 ml of 30% H_2O_2 was added to each sample and again placed on the hot plate until the sample dries. The samples were then reconstituted to 10 ml with 0.5% HNO_3 in deionized water, and filtered using Whatmann filter paper size 125 mm in a nitric acid washed 60 ml plastic bottles.

2.5 Detection of Cd in Blood Sample

Analysis of digested blood sample was carried out by atomic absorption spectrophotometry (AAS) using a Varian AA240FS with a GTA-120 graphite furnace (Varian Manufacturing Company, Australia). The instrument performed dilutions and injections automatically with a PSD 120 sampler which was fitted with tray containing 50-positions and a 2 µl dispenser. The instrument's ashing and atomization temperatures were 250 and 1800°C respectively. The spectrophotometer and auto sampler were controlled by Spectra AA version 5.1 software program. All gases used were ultrapure carrier grade and calibration curves were based on three standards (2 ppb, 4 ppb and 8 ppb). The instrument was programmed to take two readings per sample and average the absorbance. Instrument blanks (0.5% HNO_3) and check standards were processed with all samples. Sample concentrations were calculated using the final values. Concentrations of Cd in blood were determined and the results were expressed as Mean±SEM.

2.6 Accuracy and Quality Control

Accuracy of the analytical method was established through analysis of certified reference materials with index number SRM 955c (Toxic Metals in Caprine Blood) purchased from National Institute of Standards and Technology (NIST), Gaithersburg, USA. Validity of the instrument was evaluated through analysis of a known concentration of cadmium (1.50 µg/L) which was analyzed five times using the machine intended to be used for analysis of our samples and the results obtained were used to calculate precision of the machine using the following formula:

$$\frac{SD}{Mean} X100$$

2.7 Data Analysis

Mean blood cadmium concentrations of goats by location were compared using one-way Analysis of Variance (ANOVA) and Independent T-test was used to compare mean blood cadmium concentrations by sex. In the absence of reference values for blood Cd concentration in livestock, the ATSDR reference value for humans was used [66].

3. RESULTS

3.1 Analysis of Standard Reference Materials SRM 955c (Toxic Metals in Caprine Blood) for Determination of Accuracy of the Instrument

Standard Reference Material with index number SRM 955c Levels 1 and 2 was digested and analyzed together with the blood samples in order to certify the accuracy of the entire

process. Level 1 standard contains 0.0317±0.0062 µg/L while level 2 contains 5.201±0.038 µg/L cadmium in caprine blood. Following digestion and analysis, concentrations of 0.0037±0.005 µg/L and 5.225±0.003 µg/L were obtained for level 1 and 2 respectively (Table 1).

3.2 Repeated Analysis of Known Concentration of Cd for Determination of Validity of the Instrument

A solution containing 1.5 µg/L cadmium was prepared and tested five times consecutively using the machine intended to be used for analysis of cadmium in the samples. The results obtained were used to calculate precision of the instrument (Table 2). The calculated instrument's precision was 2.68% (Table 2).

3.3 Blood Cd Levels in Goats from Some Communities in Bade, Northern Yobe, Nigeria

Out of the 356 goats, 354 (99.4%) had detectable blood cadmium levels ranging from Below Detection Limit (BDL) to 27.85 µg/L out of which 349 (98.6%) had blood cadmium concentrations above the normal human reference for blood Cd concentrations set by the Agency for Toxic Substances and Disease Registry (ATSDR) (Table 3). A total of 78 (21.9%) goats were sampled from each of Lawan Musa and Sabon Gari wards, 72 (20.2%) from Katuzu, 55 (15.4%) from Dagona and 73 (20.5) from Usur/Dawayo wards (Table 3). The overall Mean±SEM blood cadmium concentration for goats in the study area was 3.998±0.21 µg/L. Goats from Dagona ward had the highest mean blood cadmium concentrations with Mean±SEM of 7.677±0.803 µg/L and the difference is statistically significant (P<0.05) when compared to blood cadmium levels of goats from all the other 4 wards. Goats from Lawan Musa, Katuzu and Usur/Dawayo wards had Mean±SEM blood cadmium concentrations of 3.996 ±0.532, 3.835 ±0.456 and 3.314 ±0.287 µg/L respectively whereas goats from Sabon Gari recorded the least mean blood cadmium concentration Mean±SEM 2.81 ±0.255 µg/L. All the goats from Sabon Gari (78), Dagona (55) and Usur/Dawayo (73) wards had blood cadmium concentrations above the normal human reference value set by ATSDR [62], whereas goats from Katuzu and Lawan Musa wards had 98.7%, and 94.8% with concentrations above the normal human reference value respectively (Table 3).

Table 1. Analysis of standard reference materials SRM 955c levels 1 and 2 for determination of accuracy of the instrument

Material	Certified value (µg/L)	Analyzed (µg/L)	Analytical Tech.
*SRM 955c 1	0.0317±0.0062	0.0037±0.005	GFAAS
*SRM 955c 2	5.201±0.038	5.225±0.003	GFAAS

**SRM 955c 1 and 2: toxic metals in caprine blood levels 1 and 2 respectively*

Table 2. Repeated analysis of known concentration of Cd for Determination of Validity of the instrument

Actual conc.	Repeated tests (µg/L)					Mean	SD	% Precision
	Test 1	Test 2	Test 3	Test 4	Test 5			
1.5 (µg/L)	1.402	1.451	1.443	1.497	1.490	1.457	0.039	2.68%

Table 3. Blood Cd levels in goats from some communities in Bade, Northern Yobe, Nigeria

Ward	Total number of goats (%)	No. of goats with detectable blood Cd (%)	Range (µg/L)	No. of goats with high* blood Cd (%)	Mean[o]±SE (µg/L)
Lawan Musa	78 (21.9)	78 (100)	0.03 – 29.32	77 (98.7)	3.996 [b] ±0.532
Katuzu	72 (20.2)	70 (97.2)	BDL – 17.85	66 (94.3)	3.835 [b] ±0.456
Sabon Gari	78 (21.9)	78 (100)	0.83 – 17.42	78 (100)	2.810 [b] ±0.255
Dagona	55 (15.4)	55 (100)	0.29 – 27.85	55 (100)	7.677 [a] ±0.803
Usur/Dawayo	73 (20.5)	73 (100)	1.04 – 17.94	73 (100)	3.314 [b] ±0.287
Total	356 (100)	354 (99.4)	BDL[a] – 27.85	349 (98.6)	3.998±0.210

[o] Levels not connected by same superscript ([a, b]) are significantly different at P<0.05
** blood Cd concentration ≥ 0.19µg/L is considered high, above the safe limit*
[a] BDL=Below Detection Limit

3.4 Comparison of Blood Cd Concentrations between Male and Female Goats from some Communities in Bade, Northern Yobe, Nigeria

A total of 91 male and 265 female goats were sampled in this study (Table 4). The mean blood cadmium concentration for the male was Mean±SEM 3.697±0.384 µg/L and that of the female was Mean±SEM 4.420±0.272 µg/L. Although the female had higher mean blood cadmium levels, statistical analysis showed no significant difference (*P*=0.299) (Table 4).

Table 4. Comparison of blood Cd concentrations between male and female goats from some communities in Bade, Northern Yobe, Nigeria

Variables	Sex	
	Male	Female
Number (n)	91	265
Mean±SEM (µg/L)	3.697^{a}±0.384	4.420^{a}±0.272
Significance level	p=0.299	

**Levels not connected by same superscript ($^{a, b}$) are significantly different at P<0.05*

4. DISCUSSION

Determination of cadmium concentrations in the SRM within the manufacturers range of ±0.03 µg/L indicates how accurate the digestion method and the instrument used for analysis of Cd in goat blood in this study are. Instrument precision of 2.67% is also indicating how close the detected values are to the real Cd concentrations in the analyzed samples. According to WHO [63], an instrument which has a Relative Standard Deviation of <10% has high precision.

The high Cd levels found in goat blood in this study strongly suggests high and extensive exposure of goats to Cd. Goats and other free ranging domestic animals are used as sentinel for environmental pollution [37,38]. This further supports the earlier findings from the area [4,5]. Presence of Cd in blood could be suggestive of long-term low-level exposure and may serve as a good reflection of the Cd body burden [64]. Animals get exposed to environmental Cd by consuming contaminated forages and water [11]. Bade area is not industrialized and no mining activities has been reported. The exact source of Cd in the area is unclear. It may probably be due to use of agricultural chemicals and chemical fertilizers [65]. There is wide scale rain-fed and irrigation farming in the area and use of chemicals and fertilizers are not uncommon. Direct input of Cd through application of phosphate fertilizers and other soil amendment products have been incriminated as one of the major sources of Cd in soil. For instance, it has been reported that Cd levels in superphosphate fertilizer may reach 46.6 mg/kg [66]. Some of the regulative focal points in many countries have being decreasing the Cd content of fertilizers and restriction on the input of Cd to farmland by application of sewage sludge and other waste products.

The widespread nature of this problem could also lends credit to the presence of high natural Cd containing ores in the area which emanates from the headwaters region in Plateau State, underlain by the basement complex rock, which is one of the main source of water to the Komadugu Yobe Basin where the study area is located [67]. Furthermore, chemicals in drainage waters from processing industries in Kano municipality which lies in the upstream part of the study area superimposed on the large- and small-scale irrigation projects in the Hadejia sub-basin may contribute to the sources of water pollution in the Komadugu Yobe Basin [67].

Sabon Gari ward, which had the least mean blood Cd concentrations in the sampled goats, is located in the Northern part of Gashua Bade local government headquarters. Goats and other animals from that part of the town usually graze in a grazing plain further north from Gashua where there is no river or major natural water body present. The area is dry throughout the year except during late rainy season when rain water from Gashua town is channeled in that direction. It is likely that the soil and grasses in Sabon Gari area are less contaminated due to limited farming activities in the area. This could possibly explain the low mean blood Cd concentration recorded from goats in Sabon Gari ward. Although not specifically determined, animals living and grazing closer to natural water bodies in this area, which is also potential source of the contamination, had higher blood Cd concentrations compared to those grazing far away from the water bodies. Patra et al. [68] reported higher blood Cd concentrations in cows grazing near a steel manufacturing plant when compared to cows grazing in an unpolluted environment.

Comparison between mean blood Cd concentrations in male and female goats showed no differences in concentration between the sexes. In humans, it was reported that women have higher Cd body burden than men [69]. Studies in cattle also suggest that females accumulate more Cd in kidneys compared to males [70]. Moreover, Doganoc, [71] and Zasadowski et al. [66] have reported that older animals accumulate more Cd in tissues than younger ones. Most of the male goats sampled in this study, particularly those from Dagona and Usur/Dawayo wards were older goats. It is probable that, the high blood Cd concentrations found in the male goats were as a result of the old age.

5. CONCLUSION

Cadmium was detected in almost all (99.4 %) the sampled goats with 93.3 % showing higher than the acceptable level and both sexes have similar exposure pattern and there was no statistically significant difference (*P*=0.299) between the blood Cd levels in male and female goats. Goats can be used as sentinels to predict environmental pollution. Exposure studies should be carried out in humans to ascertain the level of human exposure and source(s) of Cd in the studied environment need to be identified so as to control emission of this toxic metal thereby reducing exposure. Periodic evaluation of goats for Cd in the study area to give insight to environmental pollution with Cd is encouraged.

ETHICAL APPROVAL

As per international standard or university standard written ethical approval has been collected and preserved by the authors.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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