Title	Human health risks from metals and metalloid via consumption of food animals near gold mines in Tarkwa, Ghana: Estimation of the daily intakes and target hazard quotients (THQs)
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Citation	Ecotoxicology and environmental safety, 111, 160-167 https://doi.org/10.1016/j.ecoenv.2014.09.008
Issue Date	2015-01
Doc URL	http://hdl.handle.net/2115/57933
Туре	article (author version)
File Information	Ecotox.Environ.Safe_v.111.pdf



Instructions for use

- 1 Human health risks from metals and metalloid via consumption of food animals near Gold
- 2 Mines in Tarkwa, Ghana: Estimation of the daily intakes and target hazard quotients
- 3 (THQs)

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#### Abstract

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Heavy metal and metalloid contamination in food resulting from mining is of major concern due to the potential risk involved. Food consumption is the most likely route for human exposure to metals. This study was therefore to estimate the daily intake and health risk (based on target hazard quotients, THQ) from metals via consumption of free-range chicken, goat and sheep near gold mines in Tarkwa, Ghana. The concentrations of Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Cd, and Pb were measured with an inductively coupled plasmamass spectrometer and Hg analysis was done using the mercury analyzer. The mean concentrations of metals ranged from nd-542 mg/kg wet weight. Principal component analysis of the results showed a clear separation between chicken, grouped on one side, and the ruminants clustered on another side in both offal and muscle. Interestingly, As, Cd, Hg, Mn and Pb made one cluster in the offal of chicken. Chicken muscle also showed similar distribution with As, Hg and Pb clustered together. The daily intake of As (µg/kg body weight/day) were in the following ranges; [0.002 (kidneys of goat and sheep)–0.19 (chicken gizzard)], Cd [0.003 (chicken muscle)–0.55 (chicken liver)], Hg [0.002 (goat muscle)–0.29 (chicken liver)], Pb [0.01 (muscles and kidneys of goat and sheep)–0.96 (chicken gizzard)] and Mn [0.13 (goat kidney)–8.92 (sheep liver)]. From the results, daily intake of As, Cd, Hg, Pb and Mn in these food animals were low compared to the provisional tolerable daily intake guidelines. The THOs although less than one, indicated that contributions of chicken gizzard and liver to toxic metal exposure in adults and especially children could be significant.

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Keywords: Offal, Muscle, Estimated daily intake, Target hazard quotient, Ghana

#### 1. Introduction

Heavy metals and metalloid (arsenic) are ubiquitous in the environment either naturally or anthropogenically, and their concentrations were elevated promptly through waste disposal, smelter stacks, atmospheric deposition, mining, fertilizer and pesticide use and the application of sewage sludge in arable land (Cui et al., 2005). Mining and processing metal ore can be a significant source of metal and metalloid contamination of the environment (Navarro et al., 2008; Singh et al., 2005). The increased occurrence of metal and metalloid pollution in the environment has been associated with anthropogenic activities as effluents and emissions from mines and smelters often contain elevated concentrations of toxic metals including arsenic, cadmium, mercury and lead (Tubaro and Hungerford, 2007).

Food consumption has been identified as the major pathway of human exposure, accounting for >90% compared to other ways of exposure such as inhalation and dermal contact (Loutfy et al., 2006). In a review by Yabe et al. (2010), contamination of food animals, fish, soil, water, and vegetables with heavy metals has reached unprecedented levels over the past decade in some parts of Africa. As a result, human exposure to toxic metals has become a major health risk.

Tarkwa (05°18′00″N; 01°59′00″W) is a town in the southwest of Ghana, located about 120 miles west of the capital city, Accra. As of 2010, Tarkwa was estimated to have a population of 90,477 (Ghana Statistical Service, 2010). It is a noted centre of gold and manganese mining. Tarkwa mine, which is a large open-cast gold mine, is situated to the northwest of the town, and Nsuta manganese mine is situated to the east. Tarkwa has nearly a century of gold mining history and has the largest concentration of mining companies in a single district in Ghana and the West African sub-region (Akabzaa and Darimani, 2001). A study conducted by Hayford et al. (2008) on the impact of the gold mining on soil and staple foods collected around mining communities in Tarkwa showed high levels of some toxic metals including arsenic and mercury in the soil and staple foods. In Tarkwa, the extraction and processing of gold has given rise to various environmental related diseases and accidents. According to the District Medical Officer of Health, the common mining-related diseases observed in the area over the years include, but are not limited to: vector-borne diseases such as malaria, schistomiasis and onchocerciasis; respiratory tract diseases,

especially pulmonary tuberculosis and silicosis; skin diseases; eye diseases, especially acute conjunctivitis; accidents resulting from galamsey activities, and mental cases (Akabzaa and Darimani, 2001). The aim of this study was therefore to determine the concentrations of heavy metals and metalloid in offal and muscle of free-range chicken, goat and sheep; to reveal the species differences in accumulation features of metals among free-range chicken, goat and sheep; and to estimate the daily intake (EDI) and target hazard quotient (THQ) (toxic metals) through consumption of these food animals in Tarkwa.

#### 2. Materials and methods

## 2.1. Sampling and pre-treatment

In June 2012, liver, kidney and muscle samples were collected at slaughterhouses from both male and female local goat and sheep (n = 10 species each) aged three years and above, and stored in labeled plastic bags in cooler boxes. 10 free-range chickens were also bought from townships surrounding the two major gold mines. All the townships were close to the mine, and some households were within 1 km of the mine. The live adult chickens were transported to the laboratory for dissection after exsanguination. Liver, kidney, muscle and gizzard samples were collected. Samples were transported to the laboratory and kept frozen at the Chemistry Department of the Kwame Nkrumah University of Science and Technology, KNUST, Ghana before being transported and analyzed for metal concentrations at the Laboratory of Toxicology, Graduate School of Veterinary Medicine, Hokkaido University, Japan.

## 2.2. Analysis of total Mercury (Hg)

The concentration of total mercury (Hg) was measured by thermal decomposition, gold amalgamation and atomic absorption spectrophotometry (Mercury analyzer, MA-3000; Nippon Instruments Corporation, Tokyo, Japan), after preparation of the calibration standard. Recovery rates of Hg for the certified reference material, DOLT-4 (Dogfish liver, the National Research Council, Canada) ranged from 92% to 103% (94.3  $\pm$  4.2%). The detection limit (Hg) was 2.0 pg of total Hg.

## 2.3. Sample preparation and metal analysis

Approximately 0.5 g of individual samples were dried in an oven at 40  $^{0}$ C and digested in a closed microwave extraction system, Speed Wave MWS-2 microwave digestion (Berghof, Germany). Briefly, the dried samples were placed in prewashed digestion vessels followed by acid digestion using 5 mL of (65%) nitric acid, HNO<sub>3</sub> (Kanto Chemical Corp., Tokyo, Japan) and 1 mL of (30%) hydrogen peroxide, H<sub>2</sub>O<sub>2</sub> (Kanto Chemical Corp., Tokyo, Japan). The digestion vessels were capped and placed into a 10-position turntable

conditions followed by a ramped temperature programme: ramp to 160 °C (5 min hold); 119 and increase to 190 °C (15 min hold). After cooling, samples were transferred into plastic 120 tubes and diluted to a final volume of 10 mL with milli Q water. A reagent blank was 121 prepared using the same procedure. An Inductively Coupled Plasma-Mass Spectrometer 122 (ICP-MS; 7700 series, Agilent technologies, Tokyo, Japan) was used for quantification. 123 124 The instrument was calibrated using standard solutions of the respective metals (to establish standard curves before metal analysis). All chemicals and standard stock solutions were of 125 analytical-reagent grade (Wako Pure Chemicals). The detection limits (ug/L) of chromium 126 (Cr), manganese (Mn), iron (Fe), cobalt (Co), nickel (Ni), copper (Cu), zinc (Zn), arsenic 127 (As), cadmium (Cd), and lead (Pb) were 0.003, 0.025, 0.154, 0.0004, 0.024, 0.007, 0.226, 128 0.002, 0.001 and 0.001 respectively. Concentrations of metals were expressed in mg/kg wet 129 weight (mg/kg ww) 130

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- 132 *2.4. Quality assurance and quality control*
- For heavy metals, replicate blanks and the reference materials DORM-3 (Fish protein,
- the National Research Council, Canada) and DOLT-4 were used for method validation and
- quality control. Replicate analysis of these reference materials showed good accuracy
- (relative standard deviation, RSD,  $\leq$  3%) and recovery rates ranged from 80% to 115%.

- 138 2.5. Data analysis
- 2.5.1 Estimated daily intake (EDI) of toxic metals
- The estimated daily intake (EDI) of toxic metals (As, Cd, Hg, Pb and Mn) depended on
- both the metal concentration in offal or muscle and the amount of consumption of the
- respective food. The EDI of metals was determined by the following equation:

$$EDI = \frac{MC \times FDC}{BW}$$

- where MC is average concentration of metal in food (µg/g, on fresh weight basis); FDC
- 144 represents the average food daily consumption of offal and muscle in this region
- (g/person/d); BW is the average body weight. It was assumed that the local inhabitants
- 146 consumed an average liver, muscle and gizzard of 150 and 100 g/day for adult (60 kg in
- BW) and children (30 kg in BW) respectively. For kidney, it was assumed that inhabitants

148 consumed 10 and 8 g/day for adults and children respectively (Ihedioha and Okoye, 2013),

because the size of kidneys are generally smaller and is not favorite compared with liver,

muscle, or gizzard. The metal intakes were compared with the tolerable daily intakes for

metals recommended by the FAO/WHO (2010), WHO (2000) and FSA (2006).

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## 2.5.2 Target Hazard Quotient (THQ)

The health risks from consumption of offal and muscle of chicken, goat and sheep by

the local inhabitants were assessed based on the THQ. The THQ is a ratio of determined

dose of a pollutant to a reference dose level. If the ratio is less than 1, the exposed

population is unlikely to experience obvious adverse effects. The method of estimating risk

using THQ was provided in the U.S. EPA Region III risk-based concentration table

(USEPA IRIS, 2007) and in Chien et al., (2002), and is based on the equation below:

$$THQ = \frac{EFr \times ED \times FI \ R \times MC}{RfD \times BW \times AT} \times 10^{-3}$$

where THQ is target hazard quotient; EFr is exposure frequency (365 days/year); ED is

exposure duration (70 years); FIR is food ingestion rate (g/person/d); MC is average

concentration of metal in food (µg/g, on fresh weight basis); RfD is the oral reference dose

163 (mg/kg/d); BW is the average body weight, adult (60 kg); children (30 kg); AT is the

average exposure time (365 days/year × number of exposure years, assuming 70 years in

this study). Oral reference doses were based on 3E-04, 1E-03, 5E-04, 4E-03, 0.14 mg/kg/d

for As, Cd, Hg, Pb and Mn respectively (USEPA, 1997; USEPA IRIS, 2007).

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## 2.6. Statistical analysis

IBM SPSS 20.0 Statistical software (SPSS Inc., Illinois, USA) was used to perform

ANOVA analysis followed by Tukey's test. ANOVA and Tukey analyses were used to

171 compare metal concentrations in the liver, kidney, muscle and gizzard and differences were

172 considered statistically significant with p value < 0.05. Principal component analysis (PCA)

was done to determine the distribution pattern of metals in organs using JMP statistical

software v. 10 (SAS Institute).

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#### 3. Results and discussion

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- 3.1. Levels of heavy metals and metalloid
- 179 Concentrations of Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Cd, Hg, and Pb in the liver, kidney
- muscle and gizzard of free-range chickens, as well as metal and metalloid levels in liver,
- 181 kidney and muscle of goat and sheep from Tarkwa were determined (Table 1). Table 1
- shows that the mean Cd concentrations were in the range of nd-0.73 mg/kg ww in all
- samples while the mean concentrations of Pb were in the range of nd-0.39 mg/kg ww, Cr
- 184 (0.01–0.24), Mn (0.15–3.57), Fe (6.62–542.31), Co (nd–0.11), Ni (0.01–0.55), Cu (0.35–
- 185 105.8), Zn (5.12-65.89), As (nd-0.14) and Hg (nd-0.12) mg/kg ww. Cu and Zn were
- detected in 100% of all offal and muscle samples for all the food animal studied.
- Ni can cause respiratory problems and is a carcinogen. The permissible limit of Ni in
- food according to WHO (2000) standard is 0.5 mg/kg. In this study concentrations of Ni
- ranged between 0.01–0.55 mg/kg ww, in chicken liver and muscle respectively. As shown
- in Table 1, the highest mean concentration of Ni was found in the local chickens while Zn
- levels in goat muscle exceeded the permissible limit of 50 mg/kg in meat (USDA, 2006;
- 192 OJEC, 2001; European Commission Regulation, 2006).

- 3.1.1. Arsenic
- As was detectable in 100%, 83%, 80% and 52% of all gizzard, liver, kidney and muscle
- samples respectively. Concentrations of As in muscle and offal samples of free-range
- chicken, goat and sheep ranged from nd to  $0.14 \pm 0.11$  mg/kg ww in goat muscle and
- chicken kidney respectively (Table 1). The As distribution in liver, kidney and muscle of
- chicken was found to be higher and differ statistically with that of goat and sheep (p < 0.05).
- However, there was no significant difference when As levels in goat offal and muscle was
- 201 compared with sheep (Table 1, p > 0.05). Mean concentration of As in the kidney (0.14 ±
- 202 0.11 mg/kg ww) and gizzard (0.08  $\pm$  0.05 mg/kg ww) of free-range chicken from Tarkwa
- were found to be higher than the USDA (2006); OJEC (2001); European Commission
- Regulation (2006) and SAC/MOHC, 2005 standard of 0.05 mg/kg ww in food. The high
- levels of As in the free-range chicken may be due to the nature of the gold bearing ore

which is mineralized pyrites and arsenopyrates. Processing of the ore involves roasting and this results in the production of arsenic trioxide gas which is distributed throughout the study area by air current. As is a toxic substance and due to its non-biodegradable nature, it could accumulate in surface soil and water. It could also enter the food chain through plant assimilation (Amonoo-Neizer et al., 1995; Griffis et al., 2002). This could be the possible reason for the high As concentrations in free-range chickens from Tarkwa as they continuously pick food particles and water from the ground.

#### *3.1.2. Cadmium*

Mean level of Cd in chicken kidney (0.73  $\pm$  0.81 mg/kg ww) was within the recommended residual concentrations of 0.5 to 1.0 mg/kg ww in offal for human consumption (European Commission, 2001). Cd was detectable in 14% of all muscle, 100% of all liver and kidney, 90% of chicken gizzard. The concentrations of Cd in livers and kidneys of chicken were significantly higher compared to those of goat and sheep (Tukey test; p < 0.05). The higher frequencies of Cd in liver and kidney (100%) are likely due to their special functions; liver as storage organ and kidney as excretory organ (Stoyke et al., 1995). There have also been suggestions that animals exposed to Cd accumulate it in their kidneys because of the presence of free protein-thiol groups which leads to a strong fixation of heavy metals (Pompe-Gotal and Crnic, 2002). The likely unidentified sources of Cd which the animals could come in contact with include municipal waste, electroplating, plastic and paint waste. Other sources include leachates from nickel–cadmium based batteries and cadmium plated items which are so carelessly discarded by battery chargers and users in Ghana. Recently, electronic wastes are disposed and often burnt at refuse dumps.

#### *3.1.3. Mercury*

Hg is a neurotoxic poison that causes neurobehavioral effects (especially on psychomotor coordination), neuroendocrine, and renal damage and gastrointestinal toxicity (SAC/MOHC, 2005). Hg was detectable in 38% of all muscle, 69% and 97% of all liver and kidney respectively, and 100% of chicken gizzard. The levels of Hg in free-range

chicken from Tarkwa were higher compared to that of goat and sheep. The result of this study indicated that chicken kidney (0.12  $\pm$  0.08 mg/kg ww) and liver (0.11  $\pm$  0.07 mg/kg ww) contained the highest concentrations of Hg followed by sheep kidney (0.07  $\pm$  0.07 mg/kg ww). The concentrations of Hg in liver and muscle of free-range chicken from Tarkwa was found to be higher and differed statistically with that of goat and sheep (Tukey test; p < 0.05). However, there was no significant difference when Hg levels in goat offal and muscle was compared with sheep (Tukey test; p > 0.05). The lowest and highest mean concentrations of Hg in chicken were  $0.01 \pm 0.01$  mg/kg ww in muscle and  $0.12 \pm 0.08$ mg/kg ww in kidney, respectively; goat (nd-0.03  $\pm$  0.02 in muscle and kidney respectively); sheep (nd $-0.07 \pm 0.07$  in the muscle and kidney respectively) (Table 1). The USDA (2006); OJEC (2001); and European Commission Regulation (2006) permissible limit for Hg is 0.05 mg/kg and the Standardization Administration of China/Ministry of Health of China established maximum limits of Hg in food to be 0.01 to 0.05 mg/kg (SAC/MOHC, 2005). The high levels of Hg especially in free-range chicken could be problematic, as concentration levels exceeded the maximum values permitted in food. In Ghana, amalgamation is the preferred gold recovery method employed by almost all artisanal gold miners because it is a very simple, inexpensive and an easier to use technique. These activities by local and small scale miners are widespread, and Hg is introduced into the environment (via amalgamation process popularly known as "galamsey") (Amonoo-Neizer et al., 1995). The high levels of Hg in offal of free-range chicken could therefore be due to contamination from soil, feed or water.

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# 3.1.4. Lead

Excess Pb is known to reduce the cognitive development and intellectual performance in children and to increase blood pressure and cardiovascular disease incidence in adults (Commission of the European Communities, 2001). Pb was detectable in all samples of liver, kidney, 80% of chicken gizzard and 38% of muscle. The result of this study indicated that chicken gizzard (0.39  $\pm$  1.17 mg/kg ww) contained the highest concentrations of Pb followed by chicken kidney (0.26  $\pm$  0.31 mg/kg ww) and liver (0.13  $\pm$  0.14 mg/kg ww). This is because, to be toxic, Pb must be in a form which will be retained in the gizzard, thus

resulting in continuous absorption over a prolonged period (Salisbury et al., 1958). In another study by Kendall et al. (1996), analyses of field and laboratory data indicated that two scenarios can be envisioned when Pb is ingested: (1) the ingested grit/pellets containing Pb might be eliminated (via regurgitation or passage through the gastrointestinal tract) before any significant dissolution or absorption of Pb occurs; (2) the grit/pellets may be partially or totally dissolved in the gizzard, in which case causes Pb accumulation resulting in the occurrence of a range of toxicologic effects.

The levels of Pb in organs of free-range chickens may emanate mainly from contamination of soil, feeds and/ or water sources. Tukey's test was used to compare the levels of Pb in the three food animals, and results showed no statistical significance in liver and muscle when chicken was compared with goat and sheep (p > 0.05). In this study, concentration of Pb in some organs of chicken exceeded the USDA (2006); OJEC (2001); and European Commission Regulation (2006) standard of 0.5 and 0.1 mg/kg ww for Pb in offal and muscle respectively (Table 1). Generally, the low levels of Pb in the offals and muscles of the mentioned food animals could be due to the fact that, Pb accumulates in bone and the metal concentration increases with age (Caggiano et al., 2005; Rubio et al., 1998).

#### 3.1.5. Manganese

The role of Mn in neurosychiatric disorders is also documented (Schild, 1980; Jackson and Morris, 1989). Mn was detectable in 100% of all offal and muscle samples. This study showed that mean concentrations of Mn ranged from 0.15–3.51 (chicken); 0.62–2.43 (goat) and 0.71–3.57 mg/kg ww (sheep) (Table 1). The concentration of Mn in kidney of free-range chicken was found to be higher and differed statistically with that of goat and sheep (p < 0.05). However, there was no significant difference when Mn levels in the muscle of the three food animals were compared (Table 1, p > 0.05). In the liver, the levels of Mn between free-range chickens and sheep was statistically insignificant with mean values of  $3.50 \pm 0.92$  and  $3.57 \pm 1.02$  mg/kg ww respectively (Table 1, p > 0.05). The mean concentrations of Mn in offal and muscles of all three food animals were above the WHO (1996) reference standard of 0.5 mg/kg except in chicken muscle (Table 1). The high Mn

levels in these food animals could be due to the fact that the sampling site at Tarkwa was close to the Nsuta Mn mine (which is a famous site for Mn mining).

This study was compared with similar studies in different countries (Table 2). Results from this study was found to be comparable with study by Husain et al. (1996), in goat and sheep livers and kidneys, in Kuwait, but higher than similar study by Villar et al. (2005) in the liver of poultry from the Philippines. Study done by Yabe et al. (2013) near a Pb and Zn mine in Kabwe, Zambia indicated that the levels of Cd and Pb in the livers and kidneys of free-range chickens were higher compared to this study but levels in muscle were comparable. On the other hand, the levels of Pb and Cd in chicken gizzard from this study was higher than the study by Yabe et al. (2013). From Table 2, the mean concentrations of Zn in this study was comparable in all organs except for gizzard when compared to the study by Yabe et al. (2013).

# 3.2. Distribution of metals

ANOVA was used to compare the accumulation and distribution of metals in the organs of free-range chicken, goat and sheep. The metal distribution in offal and muscle of chicken was statistically significant (p < 0.05) except for Cr, As and Pb. There was also statistical significance (p < 0.01) in the distribution of Mn, Co, Cu, Zn and Cd when the organs of goat were compared. On the other hand metal distribution in organs of sheep were significant except for As (p > 0.05). The liver and kidneys are target tissues for monitoring metal contamination in animals because both organs function in removing toxic metals from the body (Husain et al., 1996; Abou-Arab, 2001). Similarly, organs of all species in this study showed that the liver and kidney contained the highest levels of metals (Table 1). Distribution of metals in offal and muscle samples in the food animals was analyzed

using PCA. PCA results (Fig. 1) showed a clear separation between chicken (C), grouped on one side, and the ruminants, goat (G), and sheep (S), clustered on another side in both offal and muscle. Interestingly, As, Cd, Hg, Mn and Pb made one cluster or group in the liver and kidney of free-range chicken (Fig. 1). Chicken muscle also showed similar distribution pattern, with As, Hg and Pb clustered together (Fig. 1). This could be attributed to the difference in their feeding habits and/ or the different levels of metallothionein (MT)

from the sampled food animals (species differences in hepatic MT content). MT is a low molecular weight protein in various mammalian and non-mammalian tissues and is involved in the detoxification of metals (Margoshes and Valle, 1957; Kagi and Vallee, 1960). According to study by Bryan-Henry et al. (1994), the hepatic MT levels in these three food animals studied decreased in order of livestock > non-mammals; i.e. goat > sheep > chicken.

# 3.3. Dietary intake of metals and target hazard quotient

The estimated daily intakes (EDI) of toxic metals for adults in the vicinity of Tarkwa mine through consumption of offal and meat of free-range chicken, goat and sheep are presented in Table 3. The highest EDIs of As (0.19 μg/kg bw/day), Pb (0.96 μg/kg bw/day), Cd (0.55 μg/kg bw/day) and Hg (0.29 μg/kg bw/day), through consumption were from the gizzard and liver of chicken (Table 3). Meanwhile, the highest EDI of Mn (8.92 μg/kg bw/day) was through consumption of sheep liver (Table 3). The calculated EDIs of As, Cd, Hg, Pb and Mn in offal and muscle samples of the 3 food animals from Tarkwa were low compared to the FAO/WHO (2010), WHO (2000) and FSA (2006) tolerable daily intakes (Table 3). Children are especially vulnerable to acute, sub-acute and chronic effects of ingestion of chemical pollutants, since they (children) consume more (twice of the amount) of food per unit of body weight as adults (ENHIS, 2007). As a result intakes of these toxic metals through food could be higher for children from Tarkwa.

The health risk from consumption of offal and muscle from the three food animals were assessed based on the THQ. THQ value less than 1 means the exposed population is unlikely to experience obvious adverse effects (USEPA, 2000). Although the calculated individual THQ were below 1.0 (Fig. 2), attention should be paid particularly to some high individual THQ values approaching 1.0 (As Cd, and Hg). The THQ for As in chicken gizzard and liver which is largely consumed was higher in children (0.85 and 0.84 respectively) than in adults (0.63 in both cases). Similarly, the THQ for Cd in the liver of chicken was 0.74 for children compared with 0.55 for adults (Fig. 2). As shown in Fig. 2, THQ for Hg in the liver of chicken was 0.76 and 0.57 in children and adults respectively. In spite of this, caution must be taken since perennial intake of these contaminated food

animals is likely to induce adverse health effects arising largely from As, Cd, Hg and Pb exposure. Meanwhile, the THQ for Mn was very low among all the three food animals (data not shown) ranging from 0.001–0.06 and 0.001–0.008 in adults and children respectively. The THQ for As, Cd, Hg and Pb showed that contributions of chicken gizzard and liver to toxic metal exposure in adults and especially children could be significant.

#### 4. Conclusion

The dietary intakes of As, Cd, Hg, Pb and Mn from the offal and muscle of free-range chicken, goat and sheep in Tarkwa, Ghana were low when compared to the FAO/WHO (2010), WHO (2000) and FSA (2006) tolerable daily intakes. Children are especially vulnerable to acute, sub-acute and chronic effects of ingestion of chemical pollutants, since they (children) consume more (twice of the amount) of food per unit of body weight as adults (ENHIS, 2007). As a result intakes of these four toxic metals through food could be higher for children from Tarkwa. THQ of As, Cd and Hg showed that there could be potential health risk for inhabitants through consumption of larger quantities of contaminated offal of free-range chicken. Prolonged consumption of these toxic metals in the offal and muscle may lead to accumulation and cause toxicity and therefore there is a clear need to avoid consumption of these contaminated food animals, as well as restrict them from roaming and scavenging for food and/or water near the mining areas. Furthermore, continuous monitoring of As, Cd, Hg and Pb residues (in these food animals) in the vicinities of gold mines in Tarkwa is recommended in the interest of consumers.

# Acknowledgement

This study was supported by a Grant-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science, and Technology of Japan awarded to M. Ishizuka (24248056, 24405004) and Y. Ikenaka (26304043) and the foundation of JSPS Core to Core Program (AA Science Platforms). We would like to acknowledge the financial support by the Mitsui & Co., Ltd. Environment Fund, The Akiyama Life Science Foundation and The Nihon Seimei Foundation. We also express our sincere gratitude and thanks to Mr Joseph Prah, Mr Joseph Addae and Mr. Takahiro Ichise who in various ways assisted to carry out this research.

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504	Figure captions:
505	
506	Fig. 1. Distribution patterns of metals in food animals characterized by PCA (C: chicken;
507	G: goat and S: sheep).
508	
509	Fig. 2. Target Hazard Quotient (THQ) of As, Cd, Hg and Pb in children and adults (C
510	chicken; G: goat; S: sheep; G: gizzard; K: kidney; L: liver and M: muscle).
511	
512	
513	

Table 1

Mean concentrations and range of metals (mg/kg ww) in various organs of free range chicken (C), goat (G) and sheep (S).

Sample		Cr	Mn	Fe	Со	Ni	Cu	Zn	As	Cd	Hg	Pb
CG(n=10)	Mean±SD	0.06±0.08	0.64±0.46	81.4±87.2	nd	0.10±0.04	1.92±0.81	30.3±3.5	0.07±0.05	0.08±0.09	0.01±0.01	0.38±1.17
	Range	0.01-0.27	0.20-1.75	37.1-321.4	nd-0.01	0.06-0.19	0.74-3.19	24.3-34.9	0.02-0.16	nd-0.30	0.01-0.03	nd-3.70
CK(n=10)	Mean±SD	0.24±0.40	2.44±0.55 a	94.4±33.2	0.03±0.01	0.07±0.14	2.82±0.37	28.4±3.1	0.14±0.11 a	0.72±0.81 a	0.12±0.08 a	0.25±0.31 a
	Range	0.01-1.35	1.65-3.59	59.7-150.5	0.01-0.07	nd-0.46	2.01-3.38	25.2-35.6	0.02-0.37	0.16-2.26	0.02-0.28	0.02-1.05
GK(n=10)	Mean±SD	0.01±0.00	0.77±0.25 b	88.8±72.8	0.07±0.03	0.14±0.07	2.89±0.46	20.0±3.9	0.01±0.01 b	0.06±0.04 b	0.03±0.02 b	0.03±0.04 b
	Range	nd-0.02	0.50-1.35	39.2-278.5	0.01-0.12	0.05-0.27	2.42-3.76	15.0-27.5	nd-0.04	0.01-0.13	0.01-0.07	0.01-0.14
SK(n=10)	Mean±SD	0.01±0.00	1.14±0.27 b	49.1±15.4	0.03±0.02	0.09±0.04	3.170±0.45	22.2±4.1	0.01±0.01 b	0.15±0.15 b	0.07±0.08 ab	0.03±0.01 b
	Range	nd-0.01	0.87-1.70	24.8-80.6	0.01-0.06	0.04-0.15	2.58-3.90	18.1-28.1	nd-0.03	0.01-0.43	nd-0.23	0.01-0.06
CL(n=10)	Mean±SD	0.02±0.01	3.50±0.92 a	542.3±604.0	0.03±0.01	0.01±0.02	3.67±0.67	65.8±50.6	0.07±0.07 a	0.22±0.18 a	0.11±0.07 a	0.13±0.14 a
	Range	nd -0.04	1.57-4.84	72.0-1629.3	0.02-0.05	nd-0.06	2.72-4.74	32.5-182.2	0.01-0.21	0.05-0.56	0.05-0.25	0.01-0.41
GL(n=10)	Mean±SD	0.02±0.01	2.42±0.71 b	89.9±47.7	0.11±0.05	0.20±0.05	60.3±44.7	47.1±26.6	nd <sup>b</sup>	0.02±0.01 b	0.01±0.01 <sup>b</sup>	0.02±0.04 b
	Range	0.01-0.04	1.06-3.41	44.3-212.6	0.04-0.21	0.15-0.29	3.3-140.7	21.9-103.3	nd-0.01	0.01-0.04	nd-0.05	0.01-0.13
SL(n=10)	Mean±SD	0.02±0.01	3.57±1.02 a	126.3±71.3	0.07±0.03	0.33±0.13	105.8±74.3	43.6±9.6	0.01±0.01 b	0.05±0.02 b	0.03±0.03 b	$0.06\pm0.03^{ab}$
	Range	0.01-0.04	1.50-4.92	55.4-294.1	0.04-0.12	0.22-0.68	7.9-224.3	31.7-63.3	0.01-0.05	0.02-0.09	nd-0.09	0.02-0.11
CM(n=10)	Mean±SD	0.05±0.01	0.14±0.08 a	6.6±1.6	nd	0.55±0.30	0.34±0.03	5.1±1.7	0.04±0.03 a	nd <sup>a</sup>	0.01±0.01 a	0.01±0.03 a
	Range	0.02-0.09	0.09-0.38	4.7-9.5	nd	0.24-1.29	0.28-0.41	3.8-10.0	nd-0.13	nd	0.01-0.02	nd-0.10
GM(n=10)	Mean±SD	0.05±0.08	0.6±0.81 a	31.7±17.0	0.02±0.05	0.14±0.05	1.50±0.76	51.3±17.6	nd <sup>b</sup>	nd <sup>a</sup>	nd <sup>b</sup>	nd <sup>a</sup>
	Range	0.01-0.28	0.21-2.83	19.2-76.8	nd-0.15	0.08-0.23	0.91-3.38	17.0-78.2	nd-0.01	nd-0.02	nd	nd-0.01
SM(n=10)	Mean±SD	0.03±0.03	0.71±0.55 a	37.8±10.9	0.01±0.00	0.46±0.16	2.09±1.19	45.9±20.8	0.01±0.01 b	0.01±0.01 a	nd <sup>b</sup>	0.01±0.00 a
	Range	0.01-0.09	0.16-2.02	22.7-58.9	nd-0.01	0.15-0.66	0.84-4.85	27.0-83.4	nd-0.02	nd-0.03	nd-0.02	nd-0.01

n: number of samples; C: chicken; G: goat; S: sheep; K: kidney; L: liver; M: muscle.

different letters (a and b) between animals in the same organ indicates significant difference (Tukey's test; p < 0.05). nd: not detected.

Table 2
Published data of mean metal levels (mg/kg dw) in organs of different animal groups from different regions.

Study/Country	Pollution Factor	Animal	Organ	Cr	Co	Ni	Cu	Zn	As	Cd	Hg	Pb
Yabe et al., 2013*, Zambia	mining	C (broiler)	L	0.06	0.05	0.03	56.80	28.60		0.00		0.06
		C(free-range)	L	0.07	0.09	0.07	3.40	30.90		1.60		4.15
			K	0.08	0.16	0.08	2.12	24.70		3.50		7.62
			Μ	0.08	0.01	0.02	nd	4.16		0.01		0.23
			G	0.10	0.04	0.03	0.13	30.50		0.02		0.23
Uluozlua et al., 2009*, Turkey	markets		L	0.04	0.02	0.01	12.10	22.50	0.06	2.24		0.12
Demirbas, 1999*, Turkey	control		L				2.95	28.10		0.04	0.08	0.09
Villar et al., 2005, Phillipines			L							0.03		0.08
Husain et al., 1996, Kuwait	wholesalers	G	L							0.05		0.13
			K							0.44		0.43
Okoye and Ugwu, 2010, Nigeria	Industrial & urban		L				134.02	120.44		0.35		0.65
Abou-Arab, 2001*, Egypt	Industrial & urban		L				46.6	32.6		0.26		0.46
	industrial & urban		K				2.4	34.1		0.91		0.68
	industrial & urban		М				1.2	41.4		0.04		0.08
Husain et al., 1996, Kuwait	wholesalers	S	L							0.04		0.13
			K							0.30		0.15
Caggiano et al., 2005, Italy	urban		L							0.33		1.50
			K							6.71		2.00
			M							0.16		1.60
Abou-Arab, 2001*, Egypt	Industrial & urban		L				48.6	36.8		0.26		0.42
			K				3	34.2		0.82		0.54

C: chicken; G: goat; S: sheep; L: liver; K: kidney; M: muscle; G: gizzard.

<sup>\*</sup> means mg/kg ww.

Table 3 Estimated Daily Intake ( $\mu g/kg$  bw/day) of As, Cd, Hg, Pb and Mn.

Sample name	As	Cd	Hg	Pb	Mn
Chicken Gizzard	0.19	0.21	0.03	0.96	1.61
Chicken Kidney	0.02	0.12	0.02	0.04	0.41
Chicken Liver	0.19	0.55	0.29	0.34	8.77
Chicken Muscle	0.12	0.003	0.03	0.03	0.37
Goat Kidney	0.002	0.01	0.005	0.01	0.13
Goat Liver	0.02	0.05	0.03	0.07	6.07
Goat Muscle	0.01	0.01	0.002	0.01	1.55
Sheep Kidney	0.002	0.03	0.01	0.01	0.19
Sheep Liver	0.04	0.13	0.07	0.14	8.92
Sheep Muscle	0.02	0.02	0.01	0.01	1.77
TDI by FAO/WHO/FSA	2.0	1.0	0.71	3.57	200

FAO: Food and Agricultural Organization.

WHO: World Health Organization.

FSA: Food Safety Agency.

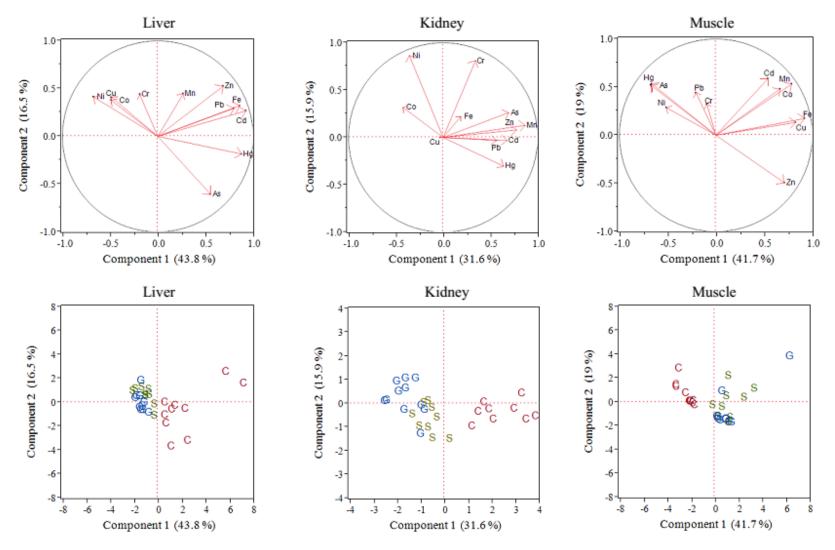
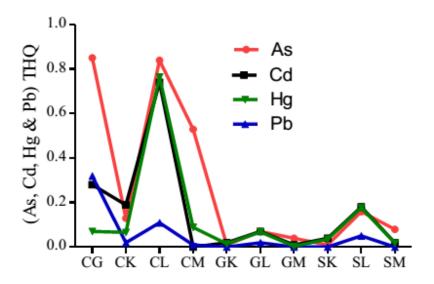


Fig. 1.

# Child



# Adult

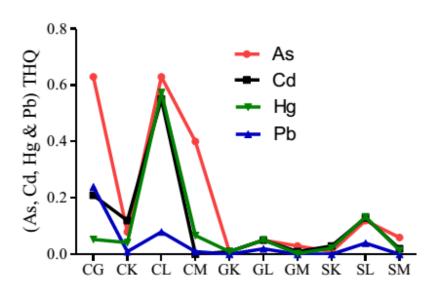


Fig. 2.