

## Food Risk Analysis Related to Heavy Metals in the Phosphates Exploitation Area of Togo

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### Abstract

Heavy metals are traces elements of natural or anthropogenic origin which present a toxic character for health and environment. Cadmium (Cd), lead (Pb) and zinc (Zn) contents were determined by atomic absorption spectrophotometry in food of selected family who practice subsistence agriculture on the phosphate mining area of Togo. Control portion was used as food study technique. The survey concerned 54 individuals (men, women and children) and revealed the presence of Pb (0.051 – 0.152mg/kg of fresh matter) and Zn (1.73 – 5.13 mg/kg of fresh matter) in the food. The proportion of individuals having a total daily dose of exposure exceeding the toxicological value of reference for Cd and Zn was estimated at 0%. The determination of essential minerals in the daily consumptions of the surveyed population showed that iron (37-109.7 mg/day) and coppers (35.6-82.9 mg/day) intake exceeded the advised nutritional values. Calcium intake (57.9-206.8 mg/day) of all the individuals was below the advised nutritional values. The food study carried out in the zone showed that the individuals of two families are at risks of toxicity related respectively to lead and cadmium. These risks can be worsened by the state of nutritional deficiencies noted in the surveyed population.

**Keywords:** Heavy metals, risk index, food as consumed, Togo

### 1. Introduction

Trace elements are metallic element present in the nature under diverse form. These traces metal elements qualified to heavy metals have diverse anthropogenic sources, like mining extractions, spreading of urban waste etc. By soil-plant or water-aquatic species transfer, heavy metals contaminate herbivorous animals and fishes, all elements of the human diet (Dube *et al.*, 2001). Due to their non-degradable nature, many living beings accumulate these toxic substances. In the body, they accumulate preferentially in the liver, kidneys and other organs such as circulatory, respiratory and nervous system (Testud, 2005) where they replace the essential elements of cell metabolism. Heavy metals are responsible for many disturbances such as anemia, cancer, immune deficiencies, diseases of muscle and skeletons and congenital anomalies (Lauwerys, 2003; ANSE, 2011). This state of affairs is often aggravated by deficient nutritional intake. The example of bone lesions in Japanese with Itai-Itai disease are the result of cadmium toxicity in combination with nutritional deficiencies, including zinc and calcium (Satarug and Moore, 2004). In Togo, after phosphate exploitation, the land are backfilled and/or restored (planting *Eucalyptus sp*, *Sennasiamea*) and use of arable land to local residents. Items often fill from excavation soil stripping phosphate minerals and gangue (consisting of non-essential elements or waste from quarries) materials. Our previous studies (Bouka *et al.*, 2012; Bouka *et al.*, 2013) in this area have revealed the contamination of soil, water, fish, vegetable and crop by cadmium, lead and zinc. The present work has been undertaken to assess the food risks related to heavy metals (cadmium, lead and zinc) in the local resident of phosphates exploitation area in Togo.

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## 2. Materials and Methods

### 2.1. Study Area

The study was conducted in 5 families who practice subsistence agriculture on the phosphate mining area of Togo. The food survey covered two groups of individual: group of adults (15 years and over) and group of children (under 15 years). The technique of the control portion was used. This technique analyzes control diets consumption of subjects for 24 hours. Information related to food preparation and consumption by individual within the household, were identified. A portion of meal (prepared on the day of the survey) consumed by each subject was weighed by a scale (Moulinex, France) and the consumption frequency of each meal was noted. Information concerning age, sex and length of stay in the area were collected. In each family, the hostess was responsible for family food cooking. The control portion of cooked dough, sauce, gari and drinking water was served and kept in ice and transported to the laboratory for analysis. The food selection was based on the eating habits of the families surveyed and especially product from their field. Local authority's (district chief) consent and informed consent from each subject were obtained before the study was undertaken.

### 2.2. Analyzes of Samples

The samples (corn dough, sauce, gari and drinking water) were analyzed for determination of cadmium (Cd), lead (Pb) and zinc(Zn) content using an Atomic Absorption Spectrophotometer (AAS 110, VARIAN) according to our previous work (Bouka *et al.*, 2012). Other trace elements in particular iron (Fe), calcium (Ca), magnesium (Mg), and copper (Cu) were determined according to the same principle (AAS).Determination of proteins was done according to standard NF V18-100. Sample organic matter was mineralized with sulfuric acid (95%)in the presence of a catalyst. Digested samples underwent Kjeldahl distillation and titration of the ammonia liberated after alkalization. Cd, Pb, Zn, Ca, Fe, Mg, Cu and protein contents in foods were expressed as mg/kg fresh weight and mg/L in drinking water.

### 2.3. Assessment of Dietary Exposure

#### 2.3.1. Estimation of Dietary Exposure

Exposure to Cd, Pb and Zn due to every food and drinking water was calculated as follows:

$$DDE = CXA / BW$$

DDE: daily dose of exposure to the contaminant; C:heavy metal concentration in food; A: total amount of food consumed per day and BW: body weight of the subject. Average adult body weights were estimated at 60 kg (Storelli, 2008). Total Daily Dose Exposure (TDDE) was calculated from the sum of different daily doses of exposure (Parent-Massin, 2009; Bonnard *et al.*, 2005) as follows:

$$TDDE = DDE_{corn} + DDE_{sauce} + DDE_{water} + DDE_{gari}$$

#### 2.3.2. Risk Characterization

Estimation of risk as an effect threshold (noncancer effects) The risk index, RI is the ratio of the daily dose exposure DDE and the corresponding toxicological reference value, TRV (Parent-Massin, 2009; AFSSA, 2007),  $RI = DDE / TRV$  RIby pollutant and cumulative RI(CRI) were calculated. RI (cumulative or not) <1, there is no adverse effect but, when  $RI > 1$  there is possibility of adverse effects.

#### 2.3.3. Nutrition Contributions

To assess the nutritional quality of food and water, the contents of Ca, Fe, Mg, Cu and protein were determined. The Daily Intake (DI) of each mineral was calculated from the formula:  $DI = CXA$  C: protein or mineral content in food and A: Total amount of food consumed per day.The total daily intake of each mineral or protein is the sum of the daily intake of various foods. DI values obtained were compared firstly to RDA (Recommended Dietary Allowance, mg / day) values to assess the risk of inadequate intake and secondarily to the safety limit (SL expressed in mg/day) to assess the risk of excessive intake of minerals (ANSE, 2011).

### 3. Results

#### 3.1. Subject Characteristics

Fifty four (54) subjects from 5 families were interviewed, including 44 adults and 10 children. The adult group (aged between 15 and 90 years) was composed of 32 men and 12 women. Their weight was estimated at 60 kg (Storelli, 2008). The children ages between 7 and 14 years. Their weight was calculated from the average weight of children per family (Table 1). Two types of meals were taken into account: 1) a full meal: corn dough with vegetable sauce which is consumed twice a day and this regularly and 2) a specific meal: gari (crushed and fried cassava tubers) taken in various forms and consumed once daily on average. The amount of food consumed per meal was calculated from the average quantities recorded in different families. The mean daily consumption in the subgroup of men was 1680 g for corn dough, 560 g for sauce and 320 g for gari. Women consumed daily 1120 g of corn dough, 470 g of sauce and 260 g of gari while in the children subgroup the values of the same meals were respectively 672 g, 260 g and 242 g. With regard to water consumption (excluding water contained in food) we assumed according to the international standard that adults and children consumed respectively 1.5 L/day and 1 L/day.

**Table 1: Subject Characteristics**

Family	Age (Year)	Weight (kg)	Number	Percentage (%)
<b>Family n°1</b>				
Man	50-93	60	2	3.8
Woman	43	60	1	1.8
<b>Family n°2</b>				
Man	17-60	60	8	15
Woman	16-44	60	3	5.5
Child	11-14	45.3	3	5.5
<b>Family n°3</b>				
Man	21-57	60	2	3.8
Woman	42	60	1	1.8
Child	8	28	1	1.8
<b>Family n°4</b>				
Man	18-65	60	6	11
Woman	16-55	60	2	3.8
Child	12	45	1	1.8
<b>Family n°5</b>				
Man	16-57	60	14	26
Woman	17-49	60	5	9.2
Child	7-12	35	5	9.2
<b>Total surveyed</b>			<b>54</b>	<b>100</b>

Adult weight was estimated at 60 kg (Storelli, 2008) and the children weight was calculated from the average weight of children per family.

#### 3.2. Content of Cd, Pb and Zn in Drinking Water and Food Preparations (Corn Dough, Sauce and Gari)

The results of trace elements determination in the food preparation and in drinking water in the different family are listed in Table 2. The concentrations of Cd in culinary preparations and in drinking water are below the detection limit ( $<6.10^{-6}$ ) excepted in family n°4 where the value reached  $0.172 \pm 0.020$  mg / kg fresh weight (fw). Lead is present with a concentration between 0.051 and 0.152 mg / kgfw. Zn content is between 1.735 and 7.152 mg / kg fw. In family n°5, the lead content in drinking water is greater than the maximum concentration recommended by WHO (0.01 mg/ mL) (WHO, 2011).

**Table 2: Content of Cd, Pb and Zn in the Drinking Water and Foods Preparation**

	Content (mg/Kg fresh matter)		
	Cadmium	Lead	Zinc
<b>Corn dough</b>			
Family n°1	ND	ND	4.875±0.174
Family n°2	ND	0.092±0.000	1.852±0.073
Family n°3	ND	ND	7.152±0.118
Family n°4	ND	0.0510±0.0005	2.622±0.060
Family n°5	ND	0.105±0.001	1.735±0.036
<b>Sauce</b>			
Family n°1	ND	ND	4.622±0.069
Family n°2	ND	ND	ND
Family n°3	ND	0.152±0.000	2.425±0.125
Family n°4	ND	0.076±0.000	2.550±0.034
Family n°5	ND	ND	4.245±0.032
<b>Gari</b>			
Family n°1	ND	ND	5.13±0.15
Family n°2	ND	ND	2.53±0.10
Family n°3	ND	ND	5.13±0.20
Family n°4	0.172±0.020	ND	3.70±0.14
Family n°5	ND	ND	3.72±0.22
<b>MCR*(mg/mL)</b>	<b>1</b>	<b>2</b>	<b>60</b>
<b>Drinking water</b>			
Family n°1	ND	ND	ND
Family n°2	ND	ND	ND
Family n°3	ND	ND	ND
Family n°4	ND	ND	ND
Family n°5	ND	0.108±0.001	ND
<b>MCR**(mg/mL)</b>	<b>0.001</b>	<b>0.01</b>	<b>0.1</b>

The results represent the mean ± SEM (n = 4)

ND: Not Determined (values are below the limit of detection for each metal considered)

MCR \*: Maximum Concentration Recommended in plants and foods (FAO / WHO, 2002)

MCR \*\*: Maximum Concentration Recommended for drinking water (WHO, 2011)

### 3.3. Dietary Exposure to Metallic Trace Elements

#### 3.3.1. Undetected Values Estimation

For the exploitation of results, undetected values were considered equal to ½ of the LD (INRA, 2004). The detection limit of Cd, Pb and Zn were respectively 0.000006, 0.00006 and 0.0010 mg/kg dw. Then undetected values were respectively for Cd, Pb and Zn, 0.000003, 0.00003 and 0.0005 mg/kg dw.

#### 3.3.2 Estimated Dietary Exposure Level

The total daily dose of exposure to Cd, Pb and Zn was reported in Tables 3. The tolerable daily intake (TDI) was taken as the toxicological reference value (TRV) according to INERIS (2009). The proportion of individuals whose TDDE exceeds the TRV established for cadmium and zinc is estimated at 0%. For lead, 44.4% of respondents (individuals of Family n°5) have a TDDE greater than the TRV established.

#### 3.3.3. Risk Characterization

Estimation of Risk as an Effect Threshold (Noncancer Effects) People of family n°4 and family n°5 have a risk index greater than 1, respectively for Cd and Pb (Table 4). The result shows that men and children from family n°3 and all individuals from families n°4 and n°5 have a CRI (cumulative risk index) greater than 1.

**Table 3: Estimated total Daily dose Exposure to Cd, Pb and Zn**

	Exposition ( $\mu$ /kg body weight/day)		
	Cadmium	Lead	Zinc
<b>Family n°1</b>			
Man	0.00019	0.00196	207.00
Woman	0.00016	0.00163	149.44
<b>Family n°2</b>			
Man	0.00019	2.57016	65.35
Woman	0.00016	1.71013	45.49
Child	0.00013	1.36016	40.99
<b>Family n°3</b>			
Man	0.00019	1.42016	250.20
Woman	0.00016	1.20013	174.74
Child	0.00022	1.41016	238.49
<b>Family n°4</b>			
Man	0.91018	2.14016	116.90
Woman	0.74015	1.55013	84.910
Child	0.92012	1.20016	73.750
<b>Family n°5</b>			
Man	0.00019	5.63016	108.00
Woman	0.00016	4.65013	91.720
Child	0.00018	5.09021	90.530
<b>VTR (effect threshold)</b>			
<b>TDI</b>	<b>0.5</b>	<b>3.6</b>	<b>300</b>

Total Daily Dose Exposure was calculated from the formula:  $TDDE = DDE_{\text{corn dough}} + DDE_{\text{sauce}} + DDE_{\text{water}} + DDE_{\text{gari}}$ , where  $DDE = \text{Content} \times \text{Amount} / \text{Body Weight}$   $TRV = \text{Toxicological Reference Value}$ ;  $TDI = \text{Tolerable Daily Intake}$  (INERIS, 2009)

**Table 4: Estimation of Risk as an Effect Threshold (Noncancer Effects)**

	Risk Index (RI)			CRI
	Cadmium	Lead	Zinc	
<b>Family n°1</b>				
Man	0.00038	0.0005	0.69	0.69
Woman	0.00032	0.0004	0.49	0.49
<b>Family n°2</b>				
Man	0.00038	0.71	0.21	0.92
Woman	0.00032	0.47	0.15	0.62
Child	0.00026	0.37	0.13	0.50
<b>Family n°3</b>				
Man	0.00038	0.40	0.83	1.23
Woman	0.00032	0.33	0.58	0.91
Child	0.00044	0.40	0.79	1.19
<b>Family n°4</b>				
Man	1.82036	0.60	0.38	2.80
Woman	1.48030	0.43	0.28	2.19
Child	1.84024	0.33	0.24	2.41
<b>Family n°5</b>				
Man	0.00038	1.56	0.36	1.92
Woman	0.00032	1.30	0.30	1.60
Child	0.00036	1.41	0.30	1.71

The risk index (RI) is the ratio of the daily dose exposure (DDE) and the corresponding toxicological reference value, TRV (Parent-Massin, 2009, AFSSA, 2007).  $RI = DDE / TRV$

### 3.4. Estimated Daily Intake of Minerals and Proteins

The values used for the RDA (recommended dietary allowance) are included in an interval where the lower limit represents the values set by AFSSA and CNRS (ANC, 2001) for children (aged 5-14 years) and the upper limit, those of adults (over 15 years). The daily intake of iron and copper exceed the RDA and safety limits reported by INRA, 2004. Calcium intake is below recommended dietary allowance (Table 5). The protein intake is lower than standard reported by Depezay, (2004).

**Tableau 5: Estimated Daily Intake of Minerals and Proteins**

	<b>Daily Intake (mg/day)</b>				
	<b>Iron</b>	<b>Calcium</b>	<b>Magnesium</b>	<b>Copper</b>	<b>Protein(g/day)</b>
<b>Family n°1</b>					
Man	109.7	182.4	564.7	65.4	7.7
Woman	86	148.7	438.3	53.7	5.9
<b>Family n°2</b>					
Man	93.4	122.4	348.5	73.8	10
Woman	74.9	95.6	262.6	58.1	7.7
Child	42.7	57.9	154.6	35.6	4.4
<b>Family n°3</b>					
Man	101.8	138.1	666.3	82.1	8
Woman	76.5	107.5	474.4	63.7	6
Child	44.1	64.7	279.3	39.1	3.4
<b>Family n°4</b>					
Man	105.8	206.8	827	74.5	5.7
Woman	77	176.1	625.1	62.1	4.1
Child	45.1	112.6	365	38.2	2.4
<b>Family n°5</b>					
Man	83.2	145.8	817.8	82.9	9.9
Woman	65.4	119.2	593.1	64.4	7.4
Child	37	70.3	347.6	39.4	2
<b>RDA</b>	<b>8-16</b>	<b>900-1200</b>	<b>200-420</b>	<b>1.2-2</b>	<b>22.5-48*</b>
<b>SL</b>	<b>28</b>	<b>2000</b>	<b>750</b>	<b>10</b>	

RDA: Recommended Dietary Allowances (ANC, 2001)

SL: Safety Limit (INRA, 2004)

\*: Protein intake in g / day (Depezay, 2004)

#### 4. Discussion

This dietary study based on the analysis of food "as consumed" was used to assess the inputs and exposure through food and drinking water in the population of the study area. Risk assessments results presented in this study take only into account this route of exposure. It is estimated that for most substances studied (cadmium, lead, zinc), food route is the primary route of exposure (INERIS, 2003; INERIS, 2005a; INERIS, 2005b). The study therefore reflects the state of inputs and exposure during the investigation, which according to international recommendations have been extrapolated to assess the long-term risks. Only the risks for non-cancer effects were calculated (literature lacks data on the cancer effects risk for cadmium and lead). It is assumed that food sampling study, consisting of a full meal and a specific food covered about 70% of daily food consumption in the area. The remaining 30% is occasional or seasonal foods consumed by this population (fruits, products such as boiled or roasted cassava bread and roasted corn). Drinking waters sampled in all families have values of Cd, Pb and Zn below the detection limit except family n°5 where Pb concentration in drinking water exceeds the value set by WHO. This is probably due to their source of supply because according to surveys, people in this family obtain drinking water from a dam located in a depression of Hahoriver, while others are supplied by well water (groundwater).

Our previous results have shown a contamination of water ponds of this area by lead (Bouka et al., 2013) and Gnandi et al, (2002) have revealed a contamination of the water of the Hahoriver by lead. All foods (corn dough, sauce and gari) have values of Cd below the detection limit except family n°4 in the case of gari. The content of metal elements (Cd, Zn and Pb) in sampled food is less than the maximum concentration recommended by FAO/WHO (2002). However, in terms of food risk management, the content of foods is not very important but the eating habits; because foods with low metal content can be toxic when consumed regularly. According to the estimation, the population of the phosphate exploitation area is exposed to cadmium, lead and zinc through the various sampled food (corn dough, sauce and drinking water). 44.4% of the surveyed population (family n°5) have daily exposure to lead (5.63 mg / kg, 4.65 mg / kg and 5.09 mg / kg respectively for men, women and children) above the toxicological reference value (3.6 mg / kg bw). This contribution is mainly due to drinking water and corn dough. 16.6% (family n°4) have daily exposure to cadmium above the toxicological reference value (0.5 mg / kg bw) due to gari. A study of the total French food done in 2011, shows a low intake of Pb (0.2 mg / kg bw / day for adults and 0.27 mg / kg bw / day in children) (ANSE, 2011).

With TDDE/TRV greater than 1, individuals of family n°5 were exposed to the toxicity risks associated with lead. Lead metal is non-essential to the human body and highly toxic even at low concentrations. 10% of Pb is absorbed by the gastrointestinal tract in adults, but the rate may reach 40% in the infant. The absorption of lead is increased in presence of fat food or during some deficiencies, particularly iron and calcium. In developing countries, adverse effects of lead are high among malnourished populations as nutritional deficiencies allow the accumulation of lead in tissues and organs (Landrigan and Todd, 1997). In other way, cadmium is a non-essential element for human metabolism, its accumulation in the body occurs mainly in the liver and kidneys (Satarug and Moore, 2004). The musculoskeletal system is not spared. Indeed, prolonged administration decreases the cadmium content of calcium in the skeleton and increases the urinary excretion (ATSDR 1999). In our study, approximately 16.6% of respondents (family n°4) have daily dose exposure to Cd greater than the TRV (0.5 mg / kg bw) doses. This contribution is mainly due to gari. According to WHO, the average dietary intake of Cd is about 10 to 80 mg/day in the normal exposure areas (WHO, 1993). A study of the diet of Canadians showed that the average daily intake of cadmium reaches about 14 mg in a range of 7-34 mg (Santé Canada, 1986). While in the United States, the average daily intake was approximately 26-51 mg (Mahaffey et al., 1975). Currently, with the emphasis on the toxic effects of Cd, this contribution tends to decrease. In France in 1998, exposure to Cd from the diet was 20 mg / day (Tremel-Schaub and Feix, 2005) against 2.7 g / day in 2004 (INRA, 2004). Uptake of cadmium at the intestinal mucosa is high in case of nutritional deficiencies in iron, calcium, vitamin D and proteins. Bone lesions in Japanese with Itai-Itai disease was the result of Cd toxicity in combination with nutritional deficiencies, including zinc and calcium (Satarug and Moore, 2004).

By combining exposure to different metals (Cd, Pb and Zn), individuals of family n°3 (7.4% of respondents) are at risk of side effects related to both lead and zinc (especially men and children). However Zn-related toxicity is very rare because zinc is an essential element in human, animals and plants. The daily iron intake (37 to 109.7 mg /day) exceeds the recommended dietary intake (8-16 mg /day) and safety limits set at 28 mg/day in all individuals surveyed. Our results are very high compared to the value (17.97 mg/day) reported in Iranian adolescents (Aberoumand, 2012). Calcium intake in all individuals (57.9 to 206.8 mg /d) is far below the recommended nutritional values (900-1200 mg /day); however our findings are still above the mean value (12.37 mg/day) of the Iranian study reported by Aberoumand in 2012. Another study has also shown a lack of calcium (451.8 mg/day) in the diet of children in rural areas of Congo (Lemonier and Ingenbleek, 1989) suggesting that calcium deficiency in diet may be common in developing countries. Calcium plays an important role in the absorption of heavy metal. It has been reported that, low calcium in diet was susceptible to provoke lead poisoning and Ca supplementation in rats decreases the absorption of Pb in the body (Meredith et al, 1977; Schields and Mitchell 1941). Calcium metabolism is also affected by the accumulation in the body of Cd which can lead to osteoporosis (Staessen et al., 1991). It was shown that magnesium were below recommended nutrient intakes in 5.5% of children and 5.5% of women and the daily intake of copper exceeded the RDA and SL reported by INRA (INRA, 2004). An intake well above those reported in the literature, which is 0.09 mg /kg bw/day in Tanzania (Ngassapa et al., 2010), 0.98 mg / day for adults and 0.81 mg / day in children less than 15 years in France (INRA, 2004). This excessive copper intake through diet in our study may result from the contamination of plants by soils. Previous work had shown a high copper content in the exploited phosphate (83-363 mg/kg) in sewage sludge (28 to 56 mg/kg) and sediment (22 to 184 mg/kg) (Gnandi, 2002).

Proteins are large molecules product by essential amino acids for the human organism and these essential amino acids must be supplied by food. The results showed that the average protein intake is below the recommended dietary intake (22.5 to 48 g / d). Deficits of 86.44% is noted in children and 67.82% in adults (men and women), while the protein needs are higher in children than in adults. A deficiency in proteins results often associated with diseases accompanying nutrients: iron and calcium (Depezay, 2004) and therefore high absorption for Cd and Pb.

## 5. Conclusion

The result of the food study in the phosphate mining area showed that individuals of family n°5 and family n°4 respectively are at the risk of toxicity associated with lead, cadmium, and these risks may be aggravated by the state of nutritional deficiencies in the area. However we recognize that this study may be an overestimated risk if we consider that the theoretical values were calculated over a period of one year without changing eating habits; and an underestimation in the sense that the casual and seasonal foods such as fruits, roasted corn, boiled cassava were not taken into account.

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