

Sources and Distribution of Mercury Residues in Environmental and Food Matrices of the Mekrou River Watershed in Kèrou, Kouandé and Péhunco in Republic of Benin

Alassane Youssao Abdou Karim^{1,*}, Mahamadou Daouda², Abdoul Kader Alassane Moussa³, Daouda Mama³, Issaka Youssao Abdou Karim⁴

¹Department of Chemical Engineering-Processes, Polytechnic School of Abomey-Calavi, University of Abomey, Calavi, Bénin

²Department of Animal Feed and Nutrition, Faculty of Agronomic Sciences, University of Abomey, Calavi, Bénin

³Department of Chemistry, Faculty of Sciences and Technologies, University of Abomey, Calavi, Benin

⁴Department of Animal Production and Health, Polytechnic School of Abomey-Calavi, University of Abomey, Calavi, Benin

Email address:

youssaoalassane@gmail.com (A. Y. A. Karim), dahouda2605@hotmail.com (M. Daouda), almokano@gmail.com (A. K. A. Moussa), mkdaouda@yahoo.fr (D. Mama), iyoussao@yahoo.fr (I. Y. A. Karim)

*Corresponding author

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Abstract: Environmental scientific research has been largely developed for about a century, and many disciplines have been interested in interactions and the role of hazardous chemical elements such as metallic mercury and its inorganic and organic compounds in the environment. This study aims to assess the mercury contamination level of some foodstuffs (water, milk) and of the terrestrial and aquatic environment (fodder, agricultural soil and sediments) in the municipalities of 2KP. Total mercury (T-Hg) has been determined by DMA-80 (Direct Mercury Analyzer) in different environmental and food matrices of the Mekrou river watershed in Kèrou, Kouandé and Péhunco (2KP municipalities). The mercury content is higher in the superficial horizon of agricultural soil (0-20 cm) than in the horizon (20-40 cm). Watercourses (Mekrou River and its tributaries) have relatively low levels of Hg (0.5 to 1.3 µg/L) in water compared with ponds and dams (181.2 to 616.9 µg/L). Conversely, ponds and dams are characterized by low levels of mercury in water and relatively higher levels in sediments. Concentrations of Hg in cow's milk and cattle feed varied respectively in the concentration ranges of (0.4 to 8.7 µg/L) and (7.1 to 15.7 µg/kg). Regarding plant material, the highest concentrations of mercury (15.7 µg/kg) are obtained in the okra leaves (*Abelmoschus esculentus* (L.) Moench) used in the human diet, compared to cattle fodder. This work reveals agriculture as the leading source of mercury ecosystem contamination, followed by Artisanal and Small-Scale Gold Exploitation (ASSGE) and hospital sources in 2KP municipalities.

Keywords: DMA-80, Pollution with Hg, Anthropogenic Sources, Kèrou, Kouandé, Péhunco

1. Introduction

Mercury (Hg), the only liquid and volatile metal in room temperature [1], is a toxic metal pollutant released into the environment by natural and anthropogenic sources [2-4]. It is mainly derived from agricultural activities (pesticides,

fertilizers), mineral exploitation and hospital sources and research laboratories ([5, 6]). These sources are not documented in the municipalities of Kèrou, Kouandé and Péhunco (2KP). However, the food needs induced by the rapid population growth have created a high agricultural production which has led to an excessive increase in pesticide imports and overexploitation of cropland. In 2007,

25365 hectares were planted and 63412 liters of pesticides used [7] against 59819 hectares and 149548 liters of pesticides in 2010 [8] in these municipalities. The increase in cultivation areas is followed by the abusive and uncontrolled use of pesticides (herbicides and fungicides), most of which comes from smuggling [9], where they are found to be prohibited or out of date. In agriculture, however, certain mercury compounds are used as antifungal agents that contribute to the release of this metal pollutant into the environment [10, 5]. In addition to the agricultural sources of Hg, we can identify Artisanal and Small Scale Gold Exploitation (ASSGE) and the use of mercury containing medical measuring instruments, in particular the mercury thermometer.

Indeed, the use of the mercury-containing medical thermometer was spread in Benin with sensitization in hospitals where each child must have his thermometer. Since mercury has the bioaccumulation capacity in organisms, its presence in the environment would lead to the contamination of the various components of the food chain and in particular humans. Among its chemical forms, methyl mercury (MeHg) is the most toxic, which has serious effects not only on the aquatic ecosystem but especially on human health. In the municipalities of 2KP, the use of pesticides for medicinal purposes, the application of pesticides with very weak and / or rudimentary protection measures, the use of the packaging of these pesticides for domestic purposes, the mismanagement of pesticides or medical waste by some hospitals and the exploitation of mining sites in a traditional way are all activities and behaviors that raise questions of research. The Decline of cotton yields and abandonment of depleted land by overexploitation of the latter and the conquest of new unexploited land are apparent consequences of the accumulation of toxic pesticide residues and mercury in particular. For all these reasons, we considered it necessary

to study the distribution of mercury residues in certain environmental and food matrices such as: surface water, sediments, cotton crop soil, cattle fodder, okra leaves (*Abelmoschus esculentus* (L.) Moench) and cow's milk in the municipalities of Kérou, Kouandé and Pehunco (2KP) located in the north of Benin. The observed levels of this metal concentration were compared to drinking water quality standards.

2. Description of Study Site

The study area of this research takes into account the watershed area of the Mekrou river in the municipalities of Kérou, Kouandé and Pehunco (2KP). It is located between 10° 00' and 11° 20' North latitude and between 1° 20' and 2° 40' East longitude (Figure 1). With a population of 289954 inhabitants and an area of 9315 km², it is limited to the North by Burkina Faso, to the South by the municipalities of Copargo and Djougou, to the West by the municipalities of Natitingou, Tanguéta and Toucountouna, in the East by the municipalities of Sinendé, Gogounou and Banikoara. Agriculture, livestock, hunting, logging, and gathering are the basis of the local economy in the municipalities of 2KP. In fact, agriculture is the first activity and the first source of income for the populations of this area. The agriculture covers about 80% of the population and cotton is the main cash crop. Livestock is the second activity of the labor force. The climate is of tropical type characterized by a rainy season which extends from April to September and a dry season going from October to March without forgetting the cold and dry wind with the haze of dust (harmattan) which blows from November to February. It is found in the municipalities of 2Kp three large permanent watercourses namely, the Mekrou, Alibori and Pendjari with torrential downpour tributaries.

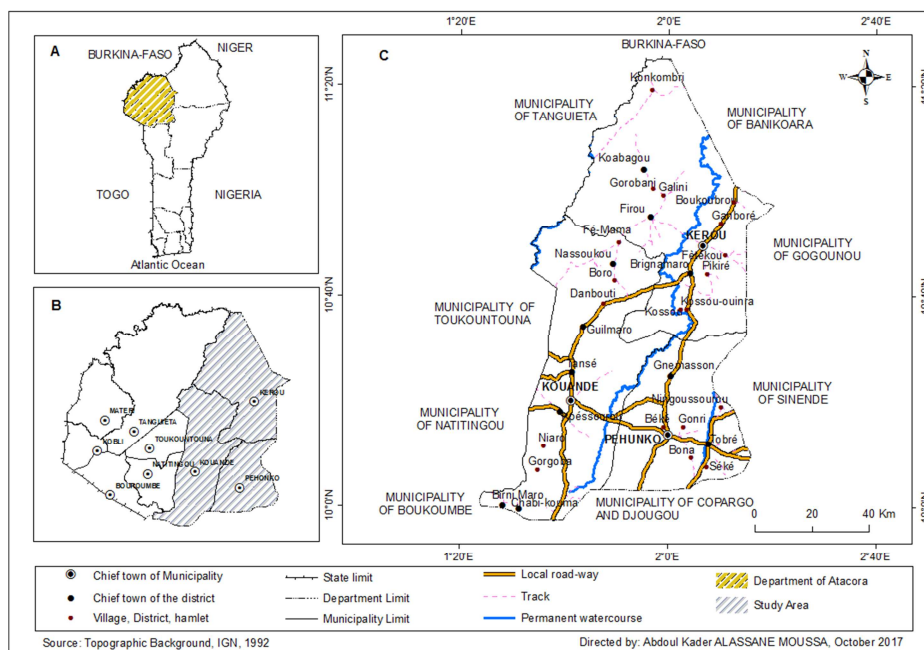


Figure 1. Geographical and administrative location of 2KP municipalities.

Figure 1. Shows the geographical and administrative location of the 2KP municipalities.

3. Materials and Methods

3.1. Collection of Study Material

For this research, the study material is composed of surface water samples (river or pond), cow's milk, sediment, cattle fodder, okra leaves (*Abelmoschus esculentus* (L.) Moench) and agricultural crop soil collected in the three municipalities around the Mekrou river. Sampling equipment (vial) was cleaned by the method called "ultra-clean" [11] which consists of cleaning with 15% nitric acid (HNO₃), 1% HCl (ACS grade reagent, JT Baker, Phillipsburg, USA) followed by seven rinses with ultrapure water (Milli-Q system; > 18 MΩ. cm) and then dried at 105°C. Agricultural soil samples were collected using dens at five locations on cotton fields, namely at the four corners of the rectangular surface and then in the middle of the field. A composite sample is made with these samples and packed in bags free of contaminants (polyethylene bag) for each of the two levels of depth i.e. 0 to 20 cm and then 20 to 40 cm (starting from the soil surface). These samples were labeled before being introduced into a cooler containing cold accumulator. The samples of fodder and okra leave (*Abelmoschus esculentus* (L.) Moench) were collected following the same processes as the agricultural soils (the four corners then in the middle of the field) and transported under the same conditions. Only forages grazed by cows were sampled by varieties, to which we added a variety grown for human consumption in association with cotton and okra leaves (*Abelmoschus esculentus* (L.) Moench).

The water samples were taken from the plastic bottles (polyethylene) previously rinsed with water from the site and then filled to the brim at about 5 cm from the surface of the body of water. These samples were acidified by adding 2ml of HNO₃ (65%) before being completely filled and sealed to prevent gas leakage. The sediment samples (river, fed and dam) were collected using a Chip peck bucket packed in bags free of contaminants (polyethylene bag) and then introduced into a cooler containing cold accumulators. (4°C).

Cow milk samples were collected from different sellers in

different Fulani camps (very early in the morning) and the composite samples stored in 0.5-liter plastic bottles free of contaminants.

3.2. Samples Preparation and Analyses

All samples are stored in the laboratory cool and analyzed directly by the MILESTONE Direct Mercury Analyzer (DMA-80) after pretreatment for solid material (soil, sediment, forage, okra leaf) and without any treatment for liquid samples (cow's milk and water). The solid samples were frozen in the laboratory at -27°C for three days. They were then weighed before being freeze-dried for 72 hours at a temperature of - 500C and a pressure of 12 bar. A sample of 200 g of each sample of sediment or soil was crushed, homogenized and then sieved using a sieve with a diameter of less than 63 µm before the analysis. As for the plant material, it is cut into small pieces before being analyzed with DMA-80 before sampling 200 g for analysis. For analysis, the samples are placed in a quartz boat on a flat scale and weighed. A mass of soil sample of 100 mg is taken in triplicate and introduced into these boats in the autosampler tray of 40 stations. DMA-80 is then started to run the analysis. This same analytical methodology was applied for samples of sediment, cow's milk, water, fodder and okra leaves.

4. Results and Discussion

4.1. Concentrations of Hg in Water and Sediments

The table below shows changes in average Hg concentrations in water in the three municipalities. It distinguishes two characteristic zones namely the weakly contaminated samples whose average concentration of Hg varies from 0.5 µg/L to 1.3 µg/L (Doh3, Pikire1, Pikire6, Kouy1, Seko2, Pikire3) corresponding to the ponds, dams and the peripheries of rivers whose water is relatively stagnant, calm and transparent, and those heavily contaminated with an average concentration of Hg ranging from 181.2 µg/L to 616.9 µg/L (Beket2, Kos4, Seko1, Kos3, Doh4, Kouy01, Makrou2) characterized by running water, high flow and disorders.

Table 1. Mean concentrations of Hg in water (µg/L) and sediments (µg/kg).

Code	Water (µg/L)	Sediment (µg/kg)	Name of river	Surface Water	characteristics
Beket2	616.9	19.2	-	River**	Plant Crops ***, troubled Water
Doh3	1.3	20	Flo	River	Periphery, calm
Doh4	297.8	40.1	Mékrou	River	Low flow, calm
Kos3	300.3	17.4	Mékrou	River	Periphery, calm
Kos4	392.3	14.8	Mékrou	River	Running water, troubled Water
Kouy1	0.8	15.5	Mékrou	River	Periphery, calm
Kouy01*	239.3	-	Mékrou	River	Running water, troubled Water
Makrou2	181.2	28.8	Mékrou	River	Running water, troubled Water
Piki1	0.9	19.9	Bouanimin	Dam	Stagnant water, calm
Piki3	0.5	24.6	Mere-Nbougnan	Pond	Stagnant water, calm

Code	Water (µg/L)	Sediment (µg/kg)	Name of river	Surface Water	characteristics
Piki6	0.9	19.6	Monri-Ndarou	Stagnant river	Stagnant water, calm
Seko1	308.2	12.4	Beaubora	Running river	Running water, troubled Water
Seko2	0.7	28.1	Werokogourou	River	Stagnant water, calm
Min	0.5	12.4	-	-	-
Max	616.9	40.1	-	-	-
Mean	180.1	21.7	-	-	-
SD	199.3	7.7	-	-	-

* Kouy01 is a site where we could not collect sediment ** - Unknown river name *** Highly developed agriculture 160 in the area. SD: standard deviation.

Caption: The sample code is composed of the three or four initials of the name of the village where the sampling site is located and an identification number (see Table 2).

Table 2. Names of Sampling Villages in 2KP Municipalities

Sample code	Beke2	Doh3, 4	Kos3, 4	kouy1	kouy01	Makrou2	Piki1, 3, 6	Seko1, 2
Village	Beket	Doh	Kossou	Kouyagou	Kouyagou	Makrougourou	Pikiré	Sekogourou

Based on the analysis of the results, sediment Hg concentrations at different sampling sites in the three municipalities ranged from 12.4 µg/kg to 40.1 µg/kg, the highest rate of Hg in the sediments being obtained at Doh (Doh4).

4.2. Concentrations of Hg in Soil Horizons, Plant Material and Cow's Milk

The figure below shows changes in average Hg concentrations in soil horizons in the three municipalities.

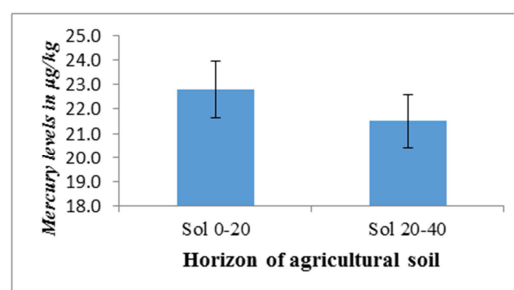


Figure 2. Variations in average Hg concentrations in soil horizons.

Table 3. Average concentrations in livestock fodder collected on cotton soils.

Espèces	HIS	IER	AEM	RCL	ACM	Min	Max	Mean	Sd
Hg (µg/L)	11.4	9.1	15.7	7.1	7.1	7.1	15.7	9.6	2.8

Based on the analysis of the results, in most cotton soil samples, mercury levels are higher (24.1 ppb) at the horizon (0-20) of the soil only in the horizon (20-40) with an average rate of 21.5 ppb.

Table 3 shows the Hg concentrations of different species of plant material collected on cotton growing soils in 2KP municipalities.

Based on the results in Table 1, four forage species and okra leaves were collected in the three municipalities: *Hyparrhenia involucrata* Stapf (HIS), *Ipomoea eriocarpa* R. Br (IER), *Abelmoschus esculentus* (L.) Moench (AEM),

Andropogon chinensis (Nees) Merr (ACM) and *Rottboellia cochinchinensis* (Lour.) WD Clayton (RCL). Hg concentrations ranged from 6.9 in RCL and ACM forage species to 15.7 µg/L in okra leaves with an average concentration of 9.6 µg/L and a standard deviation of 2.8 µg / L. the concentration of mercury is higher in okra leaves than in other forage species.

Table 4 below relates to the contamination of cow's milk with residues of Hg. The milk is collected in the peuhl's camps at Pikiré (Piki), Sékogourou (Seko), Makrougourou (Makr) and Gnemasson (Gnem).

Table 4. Comparison of Hg average in cow milk contamination rates with WHO guidelines for drinking water.

Name	Piki	Seko	Makr	Gnem	Min	Max	Mean	Sd	OMS*
Hg (µg/L)	1.1	8.7	0.8	0.4	0.4	8.7	2.8	4.0	1.0

* WHO standard.

Hg levels in milk samples ranged from 0.4 µg/L at Gnemasson to 8.7 µg/L at Sekogourou with an average of 2.4 µg/L and a standard deviation of 3.2 µg/L. The high value of the standard deviation shows a strong dispersion of the values. In addition, most of the concentrations are above the WHO standard of 1 µg/L for drinking water. The high

mercury content in cows' milk taken in Sékogourou could be explained by the high level of mercury reported in fodder and water, with cows feeding on fodder and water from the environment. All of them, according to Tables 1 to 4, the maximum values of the recorded concentrations are 8.7 ppb, 40.1 ppb, 616.9 ppb, 15.7 ppb, 34.2, ppb and 26.6 ppb

respectively in cow's milk, sediment, water, plant material and soil horizons (0-20) cm and (20-40) cm. Similarly, the average values are 2.4 ppb, 36.6 ppb, 180.1 ppb, 10.2 ppb, 22.8 ppb and 21.5 ppb, respectively. So we have the ranking in decreasing order as follows: water> sediment> soil 0-20> soil 20-40> forage> cow's milk. This classification is consistent with that of murky waters, but in relatively calm waters the concentrations in water have the lowest value

(0.85 ppb).

4.3. Distribution of Mercury in Environmental and Food Matrices in 2KP

Table 5 shows the average levels of Hg in environmental matrices and food in the municipalities of Kérou, Kouandé and Péhunco.

Table 5. Average Hg (ppb) rates in environmental matrices and foods by municipalities.

Municipalities/Matrices	Milk	Sediments	Water	Feed	Soil 0-20	Soil 20-40
Kérou	1.1	19.3	139	9.9	22.3	23.6
Kouandé	4.8	23.1	163.4	11.3	33	23.6
Péhunco	0.4	23.7	231.2	9.8	24	17.1

Referring to this table, we observe the highest concentration in Kouandé cow's milk (4.8 ppb) as well as in forage (11.3 ppb) and soil horizons (33.0 and 23.6 ppb). Hence the first source of contamination of the cow would be the agricultural source, via soil and fodder. Which gives the following classification: soil> forage> cow's milk. On the other hand, Péhunco has the highest levels in sediment (23.7 ppb) and water (231.2 ppb), followed by Kouandé. This could be explained by the presence, in the Mekrou river nest of a point source linked to the ASSGE in Pehunco at the height of Kouyagou according to the investigations results.

This study focused on the agricultural source of Hg for several reasons including the misuse of agricultural inputs for cotton cultivation. Indeed, cotton growing is by far the first in terms of planting in the municipalities of 2KP, while this crop is the most demanding in terms of use of agricultural inputs and especially herbicides and fungicides. However, Hg derivatives are used in agriculture in the formulation of herbicides and fungicides. Scientific evidence indicates that MeHg is used in agriculture as a fungicide to prevent rot [12, 13]. Mexican barley and wheat seeds that had been treated with MeHg resulted in tens of thousands of deaths among the Iraq population after consuming bread made from these products. The choice of water and sediment sampling points is made by considering the proximity of the water points of the cultivation areas and artisanal and small-scale gold mining sites [1]. More than 90% of farmers have their fields near a watercourse or body of water in the 2KP municipalities. These concerns were raised by [14] in Togo and [15] in Benin, who made reference to the nuisance that these bad practices could cause.

Levels of environmental and food matrix contamination vary from one material type to another. The concentrations in water are higher than those of WHO-free surface water (<0.5 µg/L) and WHO standards of 1 µg/L [16]. The analysis of the concentration variations reveals two trends in the water samples, the rivers with relatively higher concentrations (181.2 to 616.9 µg/L) and the ponds and dams on the other hand (0.5 and 1.3 µg/L). Variations observed in high-flow rivers and murky waters are comparable to those observed by [17] who reported 200 to 900 µg/L in the Porto Novo lagoon. In a study conducted in the Mitrovica region of Kosovo in

the Trepca and Sitnica rivers, [18] found mercury concentrations of 5 to 8.8 µg/L that are lower than the results of rivers but similar to the results of ponds and dams. [19] reported an average of 1.19 µg/L and significant seasonal variations that are close to the results of pond and dams water. In winter (1.91 µg/L), [19] found higher values than in spring (0.14 µg/L), summer (0.01 µg/L) and fall (0.01 µg/L). In all cases, all these Hg concentrations in surface water are higher than those found by [20] with rates ranging from 3 to 19 ng/L in the St. Lawrence River near Cornwall, Ontario. The heavy contamination of lake and river water in the commune of 2KP is the result of diffuse pollution from cultivated areas.

Sediment concentrations ranged from 15.5 to 81.1 µg/kg with an average of 30.1 µg/kg and a standard deviation of 23.2 µg/kg. This result is in the range of concentrations obtained by [20] in river sediments (from 15 to 882 µg/kg) but above the values reported by [21]. Soil horizons are heavily contaminated by mercury, particularly the superficial horizons (24.1 ppb) confirming permanent contact with Hg chemical pollution and its migration to the water table (21.5 ppb in the 20-40 horizon). cm). The agricultural source of Hg would be the most important for this reservoir, which is sprayed each year with tons of herbicides and fungicides [22].

Analysis results from plant material samples reveal high levels in forage and okra leaves (7.1 to 15.7 µg/L) with an average concentration of 9.6 µg/L in the range of values of literature. The total concentration of Hg reported by [20] in *N. Variegatum* ranged from 6.4 to 36.5 µg/kg. These results are also consistent with those of [22] and [19]. But [20] reported higher rates on leafy vegetables generally exceeding those of legumes and fruits [22], while [19] showed that after the roots, the leaves are the most mercury-contaminated parts. For the different species studied we have the following order of classification: AEM (15.7 µg/L)> HIS (11, 4 µg/L)> IER (9.1 µg/L)> ACM (7.1 µg/L) = RCL (7.1 µg/L). The mercury level is higher in the AEM species (Okra leaf, *Abelmoschus Eseuilleus* (L.) Moench which is a high-speed growth plant with leaves that are used for human consumption. Note that the leaves of *Abelmoschus esculentus* (L.) Moench, is grown in association with cotton. This highlights the dangers of food crops in association with

cotton and confirms the main source identified as the agricultural source. Cow's milk is the least contaminated matrix of our samples. Cattle with the ability to demethylate mercury in the rumen (rumen), beef and milk contain only very low levels of mercury [23]. But these levels are relatively high because they exceed the quality standard of drinking water. However, the presence of Hg in cow's milk is an evidence of the migration of this chemical pollutant into food webs and the exposure of populations to this pollutant through food. The classification of the average concentrations in the 2KP municipalities with water in first place of the contaminated matrices is not consistent with the results of the literature [20] that sediments are the preferred accumulations and methylation / demethylation of mercury.

5. Conclusion

The results of this study showed that all the targeted matrices showed high levels of mercury but with excessively high levels in surface water. The first source of contamination of the cow would be the agricultural source, via the soil and the fodder. This work shows the presence, in the nest of the Mekrou river, of a point source linked to the ASSGE in Péhunco at the height of Kouyagou. Agriculture and mining are mainly responsible for this pollution. However, the presence of mercury in cow's milk, okra leaves, water and in sediments deserves an in-depth study to identify the chemical forms of this pollutant in the different matrices.

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