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Herbicide Contamination in Water, Sediment and Fish *Oreochromis Niloticus* from Three Tilapia Farm in Côte d'Ivoire

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***Abstract**: The aim of this study was to determine the level of Monuron and Diuron herbicide contamination in water, sediment and farmed fish in Côte d'Ivoire. Water, sediment and fish samples were collected monthly from February to July 2017 in the tilapia cages and ponds Oreochromis niloticus from three fish farms. In these samples, residues of Monuron and Diuron were determined through high performance liquid chromatography. The results revealed that the water, sediments and fish of the farms studied are contaminated with both types of herbicides. Monuron was frequently detected in water (88%) and in fish (72%). Monthly values of Monuron range from 0.02 to 312.14 μgL^{-1} in water and from 0.02 to 303.43 μgkg^{-1} in fishes. Diuron was rarely detected in water (11%) and fish (16%). Nevertheless, it was found frequently (55%) in the sediments of all farms. Monthly values of Diuron range from 0.09 to 2.42 μgkg^{-1} in farm sediments. The average values of Diuron show no significant difference between farms. Average values of Monuron differ from one farm to another. The presence of Monuron and Diuron in the waters, sediments and fish of tilapia farms could endanger live of fishes and consumers.*

Keywords: *Agricultural activities, Côte d'Ivoire, Fish Farm, Herbicides*

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I. Introduction

In the field of agriculture, weeds are one of the major biological obstacles affecting global food production, particularly in the developing countries (FAO, 2005). In Côte d'Ivoire, the rising of agricultural activities in the 1980s led to the use of pesticides (40,000 tons) which was considered as a prerequisite to the success of a rapid agricultural development strategy, especially cash crops (coffee, cocoa, cotton, pineapple ...) (Fleischer *et al*., 1998). These pesticide products contain active molecules that are often toxic to the environment. These active substances can reach surface aquatic environments through runoff or erosion or infiltrate into the ground and reach groundwater through leaching (INRA, 2014) thus creating problem of water pollution. The pollution of the aquatic environment by micro pollutants has become a real problem to the aquaculture farms. The aquatic environment is the final receptacle to all these biocides, which can be dissolved in water, dropped on the sediments or accumulated in the food chains (Ernoult, 2009). Aquatic organisms are therefore permanently exposed to pesticide residues, some of which may persist for several years in the environment (Imorou *et al.,* 2014). Aquatic organisms living in these environments are constantly exposed. These molecules can cause, in case of large point spills, acute intoxications resulting in plants, invertebrates and fish mortality (Ernoult, 2009). The contamination of water resources via transfer phenomena is therefore a real public health problem (BRGM, 2003). Ingesting these contaminants can affect not only the productivity and reproductive capacity of aquatic organisms such as fish, but also to the health of humans for whom fish products are an important source of protein. However, in Côte d'Ivoire, Monuron and Diuron are herbicides whose utilization has long been encouraged (Silvy, 1962). Also, these two active molecules meet the criteria for ecological categorization of persistence and intrinsic toxicity for aquatic organisms. Moreover, recent studies have shown the presence of these molecules in certain Ivorian aquatic environments (Traoré *et al.,* 2015). The objective of this study was to determine the level of Monuron and Diuron contamination in the waters, sediments and fish *Oreochromis niloticus* in three tilapia farms in Côte d'Ivoire strongly influenced by antrophic activities.

II. Material and methods

2.1. Presentation of fish farms

- Fish Farm ST1 in Bingerville

The ST1 fish farm is located on the Aghien lagoon in Bingerville (South-East of Côte d'Ivoire, 18 km from Abidjan) between latitude 5 ° 24'14"N and longitude 3 ° 53'10 " W (Figure 1). It is a fish farm in the lagoon. The breeding of the tilapia *Oreochromis niloticus* is done in floating cages. Each cage has a volume of 62.5 m3 and there are about 2000 fish per cage. In the watershed area of the Aghien lagoon, there are houses, rubber plantations, palm trees and amusement sites.

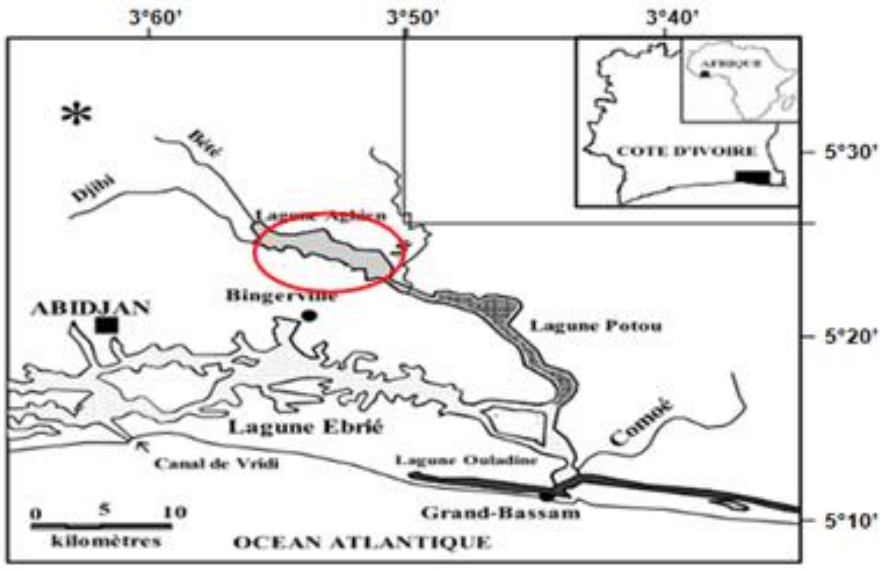

Figure 1: Fish Farm in Bingerville (ST1)

-FISH farm ST2 in Agboville

The fish farm ST2 is in a continental environment located in Offoumpo 25 km from Agboville (South-East of Côte d'Ivoire, 80 km from Abidjan) between latitude 5 ° 57'14 "N and longitude 4 ° 27 '24 "W (Figure 2). The breeding of the tilapia *Oreochromis niloticus* is in the pond. The fishing pond is a rectangular pond of 800 m^2 by 1.50 m deep. The loading density of each pond is 1fish / m^2. The farm is surrounded by rubber plantations and food crops.

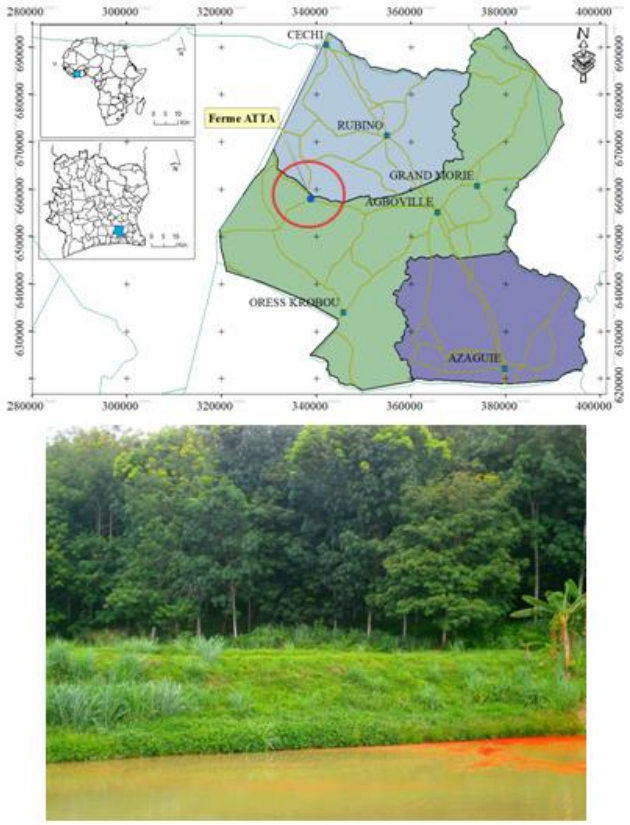

Figure 2: Fish Farm in Agboville (ST2)

- ST3 Fish Farm in Taabo

The ST3 fish farm is located on the Bandama River at the Taabo dam (Central Côte D'Ivoire, 160 km from Abidjan) between latitude 6 ° 13'32.2 N and longitude 5 ° 4'55.8 W (Figure 3). It is a fish farm in floating cages of tilapia *Oreochromis niloticus* breeding. Each cage has a volume of 62.5 m3 and there are about 2500 fish per cage. In the watershed of the farm are found plantations of coffee, cocoa and banana.

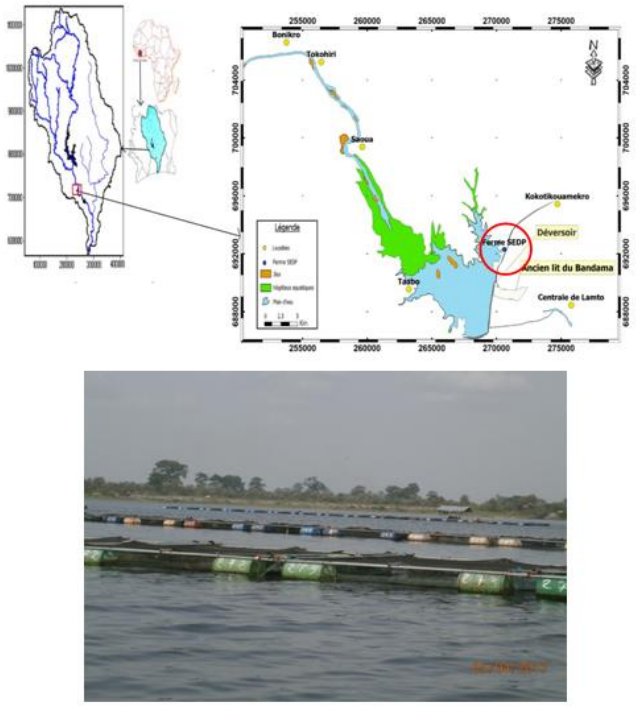

Figure 3: Fish Farm in Taabo (ST3)

2.2. Sampling of water, sediment and fish

Sampling was conducted monthly from February to July 2017 in ponds and breeding cages of *Oreochromis niloticus*. Water samples were collected using one-liter glass vials packed in aluminum foil. The sediments were collected with an aluminum tray and stored in aluminum foil. The glass vials and the aluminum tray were rinsed beforehand with demineralized water, then ultra-pure water and then 95% ethanol. The fish was sampled with gill nets and then individually wrapped in foil. All samples (water, sediment and fish) were then stored in an adiabatic chamber at 4 °C and transported to the Central Laboratory of Agrochemistry and Ecotoxicology (LCAE) for the determination of their pesticide residue concentration.

2.3. Chemical analysis and quantification of pesticide residues

The determination of Diuron and Monuron was performed through the extraction method with C-18 cartridge followed by liquid chromatography assay coupled with a mass spectrometer. Extraction and purification were done according to the recommendations of Ambrus *et al.* (1981) and Tekel and Hatrik (1996). The extracts obtained are transferred to a glass vialconic for chromatography (HPLC). The standard solutions and the samples were analyzed with a gas chromatograph equipped with a mass spectrometer, with ion scanning mode. The concentrations of Diuron and Monuron contained in the sample are calculated by comparing the peak areas of the sample products with the surfaces obtained with standard solutions of known concentrations. The expression of the results is given by the following equation:

$Cp = (Sc \times Ce \times V_2 \times Vf \times F) / (Se \times Me \times V_1)$

with:

Cp = concentration of the active ingredient (mgL^{-1}); Sc = peak area of the sample; Se = peak area of the standard; Ce = standard concentration (mgL^{-1}); V_1 = volume to be purified; V_2 = volume after purification; Vf = final volume (l); Me = volume of the sample; F = dilution factor.

2.4. Frequency of detection

The detection frequency of an active molecule is the ratio expressed, in percentage, of the number of samples where this active molecule is detected to the total number of samples taken. It is obtained from the following formula:

$FD = (Pa / P) \times 100$

with:

Pa = total number of samples containing the active molecule taken into consideration;

P = total number of samples taken.

2.5. Statistical analysis

ANOVA was used to compare the variations between farms for each of the molecules studied. Then, Tukey's Honest Significant Difference (HSD) was performed when the ANOVA showed a significant difference. The differences were considered significant at $p < 0.05$. Statistical analysis was performed using the Statistica 7.1 software.

III. Results and discussion

3.1. ***Results***

In the water samples analyzed, Monuron was detected in all the fishponds. In the ST2 fish pond and ST3, this active molecule was detected in all the samples (FD = 100%) (Figure 4). In the ST1, it was found in more than half of the analyzed samples (FD = 66%). On the other hand, Diuron was rarely detected in the ST1 and ST2 with a detection frequency of less than 20%. Diuron was not found in water samples collected from ST2 (FD = 0%). In the sediment samples collected, Diuron is the most frequently detected in all fishponds (ST1: FD = 66%, ST2: FD = 66% and ST3: FD = 33%). While, Monuron was detected only in ST1 sediment at a lower frequency (16%). Concerning the fish *Oreochromis nilotilcus*, Monuron is the active molecule that has always been detected in all fishponds as it is in the case of water samples. It was found in all the fish samples collected from the ST2 fishpond (FD = 100%). Monuron had been frequently found in the fish from ST1 at 50% and ST3 at 66%. In contrast, Diuron was detected in fish at 33% in ST1, and 16% in ST3. It was not found in the fish collected from ST2 fishpond (Figure 4).

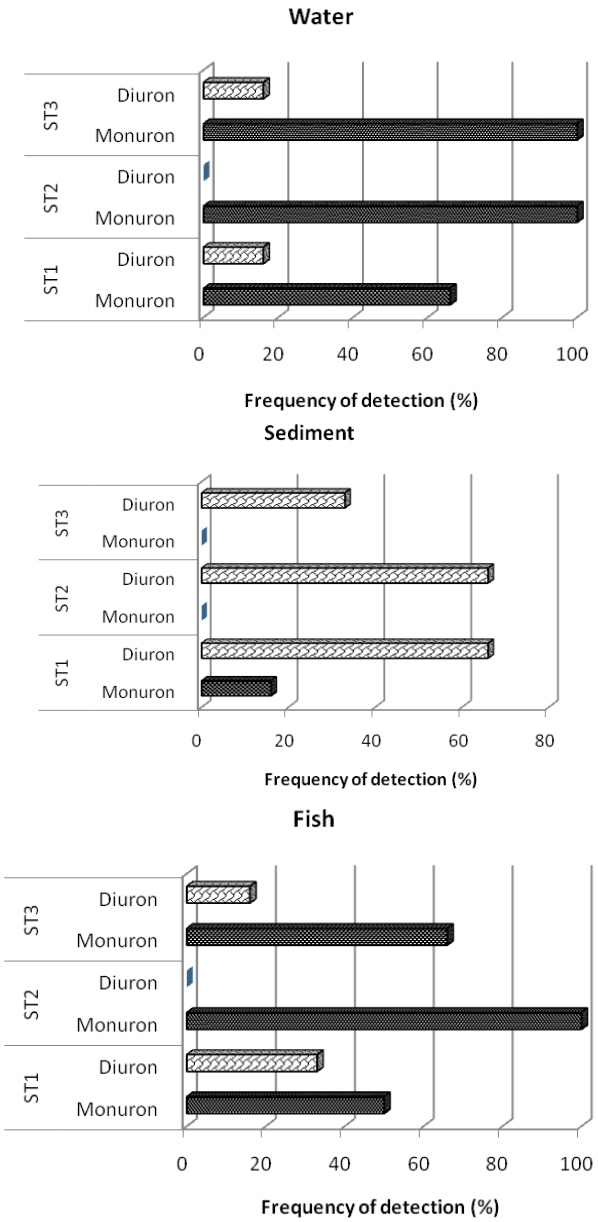

Figure 4:Frequency of detection of Monuron and Diuron in the water-sediment-fish matrix on farms ST1, ST2 and ST3

Monthly concentrations showed that Diuron ranges from 1.55 to 1.77 μgL^{-1} in water, 0.09 to 2.42 $\mu g\ kg^{-1}$ in sediment and 0.28 to 3.50 μgkg^{-1} in the fish. In contrast, a monthly concentration of Monuron goes from lower concentration in the sediments to higher concentration in the water and fish (Table 1). Monuron ranges monthly from 0.02 to 312.22 μgL^{-1} in water and from 0.02 to 303.43 μgkg^{-1} in fish. It was not generally found in the sediment, only one value was noted (1.53 μgkg^{-1}) in the ST1 fishpond.

The average values of Monuron showed a significant difference, when compared ($p < 0.05$) between the fishponds under observation, in water and in the fish. They are higher in the water of ST2 fishpond and in the fish in the other two fishponds. However, the mean concentrations value of Diuron showed no significant difference when compared ($p > 0.05$) between the farms studied in water, sediment and in the fish (Table 2).

***Table 1*:** Monthly minimum and maximum values in water (µg/L), sediment (µg/kg) and fish (µg/kg) from fish farms ST1, ST2 and ST3

		Fish farms					
		ST1		**ST2**		**ST3**	
Herbicides	**Matrix**	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Monuron	**Water**	2.19	2.47	0.02	312.22	0.13	35.26
	Sediment		1.53	ND	ND	ND	ND
	Fish	4.03	303.43	0.02	50.36	32.54	112.54
Diuron	**Water**		1.55	ND	ND	ND	1.77
	Sediment	1.05	2.42	0.09	0.23	0.66	0.97
	Fish	0.46	3.50	ND	ND	ND	0.28

***Table 2*:** Mean values in water (µg/L), sediment (µg/kg) and fish (µg/kg) from fish farms ST1, ST2 and ST3

		Fish farms					
		ST1		**ST2**		**ST3**	
Herbicides	Matrix	Average	S-D	Average	S-D	Average	S-D
Monuron	**Water**	1.56^{a}	1.21	145.15^{c}	158.24	17.46^{b}	18.09
	Sediment	0.25	0.62	ND	-	ND	ND
	Fish	64.63^{b}	1.33	21.84^{a}	24.37	51.84^{b}	48.01
Diuron	**Water**	0.25	0.63	ND	-	0.29	0.72
	Sediment	1.10	1.00	0.11	0.10	0.27	0.43
	Fish	0.66	1.40	ND	-	0.04	0.11

Values with letters a, b, c in superscript show a significant difference ($p < 0.05$) between farms, S-D: Standard deviation

3.2. Discussion

Our results showed the detection of Monuron and Diuron in the water-sediment-fish matrix in the studied fish farms. The presence of these active molecules in all the compartments of the studied fish farms (water, sediments and fish) is due to the physicochemical properties of the matrices (pH, temperature) (Lalancette, 2012). Moreover, the physicochemical properties (solubility, half-life time, polarity) of the active ingredients also come into play. Indeed, Desgranges (2015) stated that the characteristics of pesticides multiply their possibilities of contamination of the various compartments of the environment. In addition, Diuron and Monuron are a group of herbicides that are quite soluble in water and lasting in the sediments (Ramade 2011). The high frequency of Monuron in water can be explained by the fact that water is the first ultimate receptacle for pollutants in an aquatic ecosystem (Lazartigues, 2010). According to Guigon-Moreau (2006), pesticides are leached in dissolved form and associated with suspended particles to surface water before degrading or migrating to sediments or accumulating in aquatic organisms. However, the high prevalence of Monuron in both fish and water is due to the fact that aquatic organisms accumulate active molecules of pesticides from water through gills and epithelial tissue (bio concentration), but also from food (biomagnification) (Agbohessi *et al.*, 2012). The results of the detection frequency also showed the high presence of Diuron in the sediments. This could be explained due to the fact that Diuron is easily absorbed by sediments in which it slowly degrades biologically (Andral, 1996). Sediments are also the receptacles for pollutants which could remain trapped for months or even years (Diop, 2013). These two active molecules frequently found during this study were marked by the European Union because they meet the criteria of ecological categorization of persistence and intrinsic toxicity for aquatic organisms. In addition, they have been shown to be mutagenic and teratogenic (Tron *et al*., 2001).

Monthly concentrations have shown that Diuron has low values (from 1.55 to 1.77 μgL^{-1} in water, from 0.09 to 2.42 μgkg^{-1} in the sediments and from 0.28 to 3, 50 μgkg^{-1} in fish) in all compartments compared to Monuron, which often has critical values (303.43 μgkg^{-1} in fish and 312.22 μgL^{-1} in water) dangerous to the health of ecosystems. These often very high monthly values of Monuron suggest recent pollution of farm water probably due to significant runoff just after the application of phytosanitary products (Diop, 2013). In addition, Monuron has a very high solubility 200 mgL^{-1} (Boucheloukh, 2013). This recent pollution shows that Monuron is still circulating in the Ivorian markets, whereas it has been banned by the European Union since 1994. It is either always authorized by the Ivorian Government or fraudulently acquired (M.A., 2014). These high values of Monuron can have effects on fish growth and reproduction. However, the behavior of Diuron in this study shows that it is an old pollution probably due to the long lasting effect of this molecule.

The average values of Monuron in water and fish showed a significant difference between the farms studied. This difference between farms is probably related, on one hand, to variations in the physicochemical parameters of farm water, to site structures, pond and cage areas, and on the other hand, to the distance from the farm application site of the pesticides to the water source. Indeed, the fishpond (ST2) is surrounded by rubber plantations whose distance does not exceed 20 m. This would explain its high pesticide contamination compared

to other farms whose plantations are located in the catchment area of the streams that house the fishpond. According to Añasco *et al*. (2010), the most contaminated surface waters by pesticides are those located at sites closer to agricultural land.

The concentrations of Monuron (0.02 to 312.22 μgL^{-1}) obtained in farm waters in this study are higher than those reported (0.00-52.95 μgL^{-1}) by Traoré *et al*. , 2015 in the Aghien lagoon. However, our results are similar to those of Martin *et al*., 2013. These authors detected Diuron in sediment four years after it has been banned.

Our values of Diuron in water (1.55 to 1.77 μgL^{-1}) and in sediments (0.09 to 2.42 μgKg^{-1}) are higher than those obtained by Sarangaraja *et al.,* 2012 in the same matrices (0.01-0.0 62 μgL^{-1} in water and 0.01-0.09 μgg^{-1} in sediments) in Japan. The different climatic conditions of the countries could explain these differences.

IV. Conclusion

This study showed the presence of Diuron and Monuron in the waters, sediments and the tilapia fish *Oreochromis niloticus* from the fishpond of Aghien (ST1), Offumpo (ST2) and Taabo (ST3). The application of phytosanitary products and the runoff from rubber trees and food crops along the farms are believed to be responsible for this pesticide contamination. The detection of these active substances already banned in Europe by the European Commission of Agriculture and Rural Development due to their toxicity or high persistence in the environment constitutes a real danger of toxicity to farmed fish and to consumers alike.

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