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Occurrence and Risk Assessment of Pyrethroids and Organophosphorus Pesticides in the Middle Part of Mono River Basin

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Abstract: Pyrethroid and organophosphate insecticides are globally recognized as the most extensively used insecticides. Their potential to accumulate in groundwater raises significant concerns due to the severe threats they pose to both human health and the environment. This study examined the presence of organophosphorus and pyrethroid pesticides in rivers in the middle of the Mono river basin and their ecotoxicological risk. To reach these aims, seven organophosphorus pesticides and three pyrethroids were investigated in thirteen water samples by liquid-liquid extraction and GC-MS/MS analysis. The ecotoxicological risk was assessed for fish and crustaceans using the risk quotient method, which is the ratio between the measured environmental concentration and the toxic reference value. The results show that all the targeted pesticides were found, with maximum concentrations of 0.9647; 0.8947; 0.0978; 0.0908; 0.0602; 0.0578; 0.0483; 0.0468; 0. 0245 and 0.0152 $\mu g.L^{-1}$ respectively for Lambda-cyhalothrin; Chlorpyriphos; Deltamethrin; Fenchlorphos; Cypermethrin; Prothiofos; Dichlorvos; Ethoprophos; Disulfoton and Parathion-Methyl. The detection rate for the first two pesticides was 100%, and 61.54% of water samples had concentrations above 0.5 $\mu g.L^{-1}$, which is the threshold limit for pesticides in drinking water. At the sampling points, the concentration increases with the distance travelled by the rivers. Risk assessment for aquatic species shows negligible risk for fish in most rivers analysed. For crustaceans, the risk quotient reveals a moderate and low risk for 46.16% and 53.84% of the rivers analysed respectively.

Keywords: Pesticides, Mono River Basin, Ecotoxicological Risk, Water Pollution

1. Introduction

Agriculture is one of the economic «engines» in developing countries. The appearance of agricultural pests has forced farmers to use pesticides to limit yield losses [1] for over fifty years, and pesticides use increases more and more each year. However, several studies have shown that only a small proportion of these pesticides reach their targets after application; the rest end up in the environment notably in surface water, soil, and air [2]. This accidental pollution has a considerable negative impact on the environment. In water, some of these pesticides can modify the DNA structure of aquatic communities [3]. Pesticides are constantly evolving, with new formulations emerging all the time. They are mainly characterized by their diversity, different chemical properties, and low concentrations in real samples [4]. Organochlorine family are the main family of pesticides uses in the past. Due to their harmful effect, these compounds are replaced by organophosphate, pyrethroid, triazine and other groups. Organochlorines are mainly banned due to their persistence in environment leading to a strong use of new molecules which

are, then among the most used pesticides detected in surface water worldwide [5]. Pyrethroids are the latest generation of pesticides, highly stable to light and temperature, although they can undergo rapid biological degradation in the environment.

In West Africa, and Togo in particular, agriculture is a top priority because of it weigh in Gross domestic product (GDP). It is also heavily dependent on pesticides. Pyrethroids (PYR), organophosphorus (OP), triazine and carbamate pesticides are the most widely used families throughout the country [6, 7]. Although these groups of pesticides are considered easily biodegradable, some are extremely toxic, especially to aquatic organisms.

Unfortunately, few studies have been carried out on the pollution status of this family of pesticides in Togo, particularly in the Mono River basin where fishing is highly practiced. This study trends to fill the gap, with the aim of assessing the pollution status of OPP and PYR pesticides in waters rivers in the middle Mono river basin with a view to identifying the risk on aquatic organisms. This study is necessary for this basin because, despite its socio-economic importance for Togo, few studies have focused on the quality of these waters.

2. Methodology

2.1. Description of the Study Area

The Mono River basin stretches between latitudes 6°16' and 9°21'N, and longitudes 0°42' and 1°55'E with an area of around 24,100 km^2 (*Figure 1).* This basin is shared by Togo and Benin in its southern part, while its central and northern parts are entirely in Togo. Two climatic domains are distinct in the Mono basin: a Sudanian climatic domain with a unimodal rainfall regime (May to October) in the upper part, and a Guinean climatic domain classified as subequatorial with a bimodal rainfall regime (April to July and September to November) in the lower part [8]. The central part of the basin represents the transition zone between these two climatic conditions. Annual rainfall varies from 1,500 mm to less than 1,000 mm. The middle part of the basin is home to the Nangbeto dam, used for power generation and fishing. This part, characterized by numerous human settlements, is one of the country's main cereal and cotton production zones [9] and regularly hosts transhumant livestock [10].

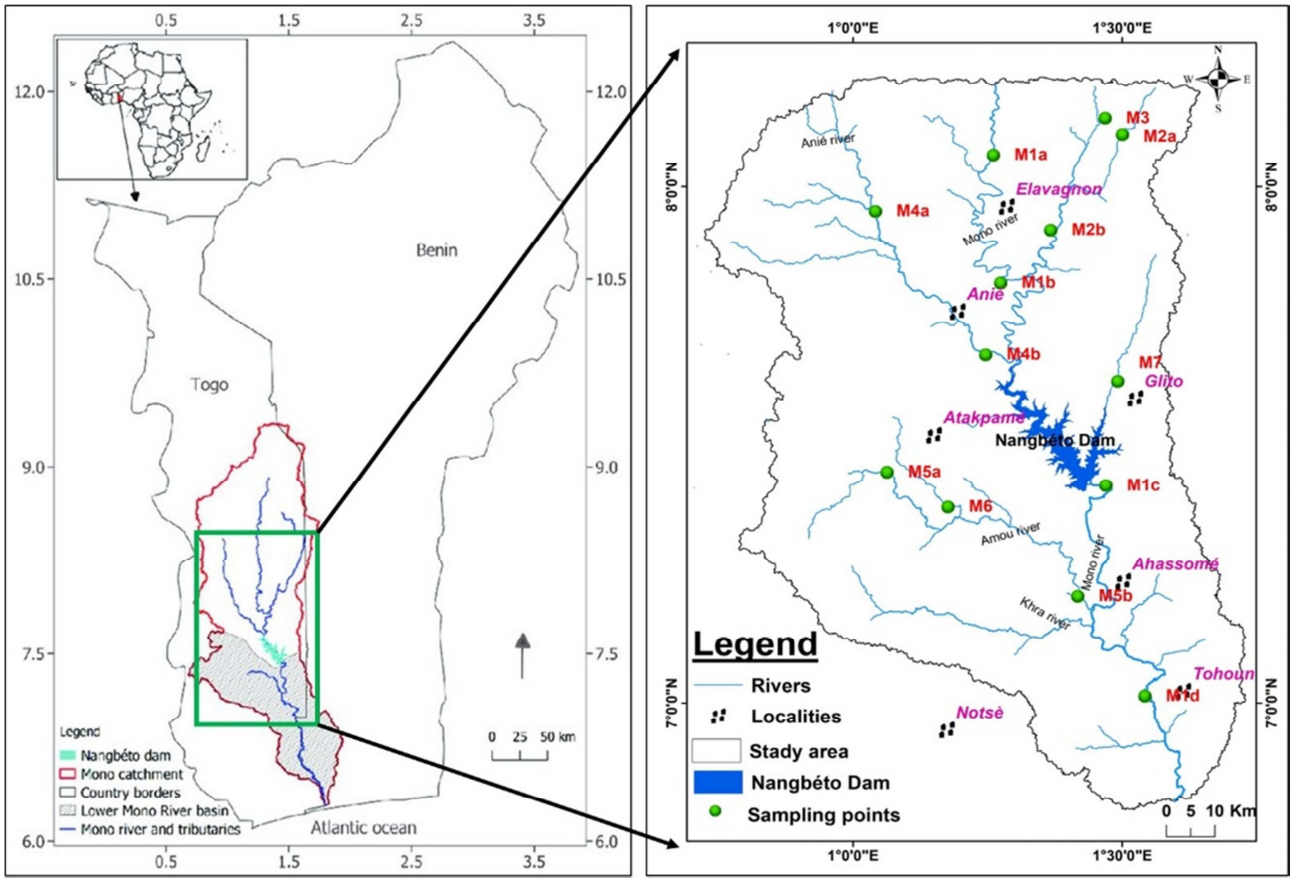


Figure 1. *Map of mono basin environment and sampling points.*

2.2. Sample Collection

A total of 13 water samples *(Figure 1*) were collected in July 2023 during rainy season along the Mono River and its major tributaries. These water sources are used by the local inhabitants for drinking, bathing, and fish farming. Some are used to supply large cities with drinking water. The water samples were collected in one-litter glass bottles, with a screw cap fitted with a Teflon-coated seal, previously washed with water and ethanol, rinsed with acetone, and dried at 120°C for 2 hours. Samples are covered with aluminium foil, kept on ice

(0°C) during transport and then stored at 4°C in the refrigerator until analysis.

2.3. Reagent and Solvent

The standards of pesticides, the internal standards (Cis-cypermetrine-d5 and Triphenyl phosphate TPP) with the 99% purity, NaCl, anhydrous sodium sulphate, dichloromethane (DCM), hexane and acetone analytical grade were purchased from Sigma Aldrich (St-Quentin Fallavier, France).

The 10 pesticides evaluated belong to the pyrethroid family and the organophosphorus family whose chemical structures are illustrated in *Figure 2*. The target pesticides are greatly use in Togo and acute toxicity is well known.

2.4. Extraction

Pesticide residues in water samples were extracted by liquid-liquid extraction (LLE) with Dichloromethane. Briefly, in a separatory funnel, 0.5L of unfiltered water sample was introduced, followed by pesticides standard and then 80mL of extraction solvent. The mixture was stirred for 15 min, while occasionally releasing the pressure and left to settle for phase separation. The organic layers were separated from the aqueous layers. The aqueous phase to be decanted was taken and extracted twice more with 50mL of the solvent mixture. The combined organic extracts from each sample were filtered twice over a bed of anhydrous sodium sulphate to remove residual water, and concentrated to dryness using a rotavator at temperatures between 35 and 40°C. The dried extract was then recovered with 2 ml hexane, 0.250 mL of internal standard was added (Cis permethrin d5 for Pyrethroids and TPP for organophosphorus) and further concentrated under inert nitrogen to 250µL before injection into GC-MS/MS.

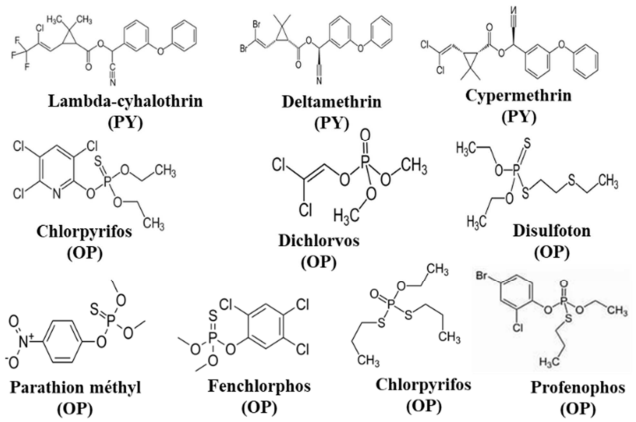


Figure 2. *Structure of Target pesticides.*

2.5. GC-MS/MS Analysis

Pesticides were determined by an Agilent 7890 B GC with an Agilent 7000C Triple Quadrupole GC/MS system. The GC was equipped with an Agilent DB-17ms column (30 m × 0.25 mm, 0.25 µm) and helium (with a purity of 99.999%) served as the carrier gas, flowing at a constant rate of 1.2 mL/min. 1 µL of the extract was injected for analysis.

The oven temperature was set to 70°C for 2 min, then increased to 150 °C at a rate of 25 °C/min, after to 200°C at a rate of 3°C/min and finally to 280°C at the rate of 8°C/min, with a hold time of 1.5 min. The mass spectrometer was run using electron impact ionization (EI) at 70 eV energy and in multiple reaction monitoring (MRM) mode for a high selectivity. Qualitative and quantitative interpretation was performed by Agilent MassHunter software.

2.6. Method Validation

A method validation study was carried out first. Water samples were spiked with a solution containing a mixture of the desired pesticide standards. Blank and recovery experiments were carried out, evaluating recovery rate, linearity, limit of detection and limit of quantification. Water spiked with the desired pesticides (1 to 1000 ppb) was extracted and analysed following the above protocol. The limit of detection (LOD) was obtained for a signal-to-noise ratio equal to three (S/N =3) and the limit of quantification (LOQ) for a signal-to-noise ratio equal to ten (S/N =10). Data were not corrected for recovery rate.

2.7. Ecological Risk Assessment

There are many approaches for health risk assessment exposure to various mixtures of chemicals.

In this work, the ecotoxicological risk was assessed using the European Food Safety Authority (EFSA) risk quotient (RQ) method for fish and crustacean in water, as described by Bai [11]. Each pesticide risk quotient (RQ) was quantified for using the ratio between the measured environmental concentration (MECi) and the toxic reference value (PNEC) (Equation 1).

$$\mathrm{RQ} = \sum_{i=1}^{n} \mathrm{RQi} = \sum_{i=1}^{n} \frac{MECi}{\mathrm{PNECi}} \qquad (1)$$

The PNEC is the predicted no-effect concentrations obtained by dividing the 96 h-LC_{50} (half lethal concentration for the 50% of the population) or the NOEC (No observation effect concentration) by assessment factor (AF =1000). The choice of 1000 as an assessment factor is conservative but is based on absence of chronic toxicity data) of pesticide. For most pesticides, LC_{50} and NOEC values were obtained from the Pesticides Properties Database (PPBD) show in *Table 1*

Furthermore, previous studies based on no synergistic or antagonistic risks for pesticides [12, 13], suggested that the combination effects of total pesticides risk (RQ) in water samples can be calculated by the sum of their individual RQi (Equation 1).

Risk was characterized by the RQ values as follow: when RQ exceeds 1, it indicates high risk; 0.1 < RQ < 1, moderate risk; 0.01 < RQ < 0.1, low risk; and RQ < 0.01, negligible risk [14, 15].

Table 1. *Toxicity data of the eight target compounds to the aquatic species [16].*

Pesticide	Fish ((µg.L^{-1})			Crustacean (µg.L^{-1})	
	LC50	NOEC	PNEC	LC50	PNEC
Chlorpyriphos	25	0,14	14	0,04	4
Dichlorvos	550	110	11000	19	1900
Disulfoton	39	220	22000	100	10000
Ethoprophos	320	64	6400	15	1500
Fenchlorphos	740	-	74000	-	0
Parathion-Methyl	270	8,9	890	0,31	31
Prothiofos	500	-	50000	-	0
Cypermethrin	1,51	0,03	1510	12,8	12800
Lambda-cyhalothrin	0,21	0,031	210	0,003	3
Deltamethrin	0,15	0,032	150	0,0017	1,7

Table 2. *Analytical performance of target pesticides.*

Pesticides	Recovery (%)	Linearity (R^2)	LOD (µg.L^{-1})	LOQ (µg.L^{-1})
Chlorpyriphos	91 ± 8	0.9960	0.0025	0.005
Dichlorvos	86 ± 15	0.9938	0.0030	0.005
Disulfoton	73 ± 3	0.9933	0.0010	0.005
Ethoprophos	76 ± 12	0.9973	0.0010	0.005
Fenchlorphos	75 ± 7	0.9953	0.0010	0.005
Parathion-Methyl	70 ± 12	0.9981	0.0010	0.003
Prothiofos	75 ± 6	0.9994	0.0010	0.005
Cypermethrin	79 ± 9	0.9994	0.0025	0.005
Lambda cyhalothrin	89 ± 8	0.9986	0.0025	0.005
Deltamethrin.	89 ± 11	0.9921	0.0010	0.003

Table 3. *Pesticide concentration and detection frequency.*

Variable	Frequency (%)	Min (µg.L^{-1})	Max (µg.L^{-1})	Mean (µg.L^{-1})	Std. deviation
Chlorpyriphos	100	0.0239	0.8947	0.4523	0.3321
Dichlorvos	62	Nd	0.0483	0.0161	0.0174
Disulfoton	31	Nd	0.0245	0.0047	0.0082
Ethoprophos	31	Nd	0.0468	0.0075	0.0155
Fenchlorphos	15	Nd	0.0908	0.0078	0.0251
Parathion-Methyl	8	Nd	0.0152	0.0012	0.0042
Prothiofos	15	Nd	0.0578	0.0068	0.0175
Cypermethrin	85	Nd	0.0602	0.0267	0.0181
Lambda-cyhalothrin	100	0.009	0.9647	0.4030	0.3604
Deltamethrin	69	Nd	0.0978	0.0122	0.0260

3. Results and Discussion

3.1. Analytical Performance

The result of analytical methods validation was detailed in the *Table 2*. The limit of detection ranges from 0.001 to 0.01 µg/L and the limit of quantification range from 0.005 to 0.02 µg/L. The method shows a good recovery varied from 73±3% to 91±8%. Amount the linearity; regression gave a good R^2 higher than 0.99. All these factors indicate that this method is acceptable for target pesticides assessment in water.

3.2. Detected Pesticides

OPP and PYR pesticides are widely used due to their ready availability, wide range of efficacy and ability to control a large pest. In our study area, the ten targeted insecticides were detected and summarized in *Table 3*. Two pesticides (Lambda-cyhalothrin and chlorpyrifos) were present in all samples and were also the insecticides with the highest concentrations, i.e., 0.9647 µg.L^{-1} and 0.8947 µg.L^{-1} respectively, despite their low solubility in water. Similarly, cypermethrin, dichlorvos and deltamethrin have a frequency of occurrence of over 50%. Studies carried out in Togo pointed out the abusive and uncontrolled use of pesticides, especially chlorpyrifos [17], lambda-cyhalothrin and cypermethrin [6, 18], in the vicinity of rivers and market gardening due to their relative availability on market.

Comparing maximum concentrations of WHO guidelines for drinking water of 0.1 µg.L^{-1} per individual substance [19], chlorpyrifos and lambda-cyhalothrin exceed this value.

3.3. Distribution of Pesticides

The contamination status per sample is presented in *Figure 3*. More than 60% of waters samples were characterized with a sum of pesticides above the WHO standard (0.5 µg.L^{-1}) in drinking water [19, 20].

The sample M1a, M1b, M1c and M1d are from the Mono River, the main river in the basin (*Figure 1*). The pesticides amounts are among the highest probably due to addition effect. Indeed, pesticides found in these samples could come from up stream practices in addition to local practices in the study area.

The maximum concentration (*Figure 3*) of this river is obtained at the outlet of the Mono dam (M1C), due to the strong agricultural practices in this part of the Mono dam and the significant input of countless tributaries. The fish farming practiced in this area also reveals a variety of chemicals. In fact, all the targeted pesticides were detected in this sample. The pesticides content decreases as we move away from this area, despite the contribution of other effluents. This may be due to adsorption capacity of pesticide on sediments and degradation processes. The samples from this river were mainly polluted by chlorpyrifos and lambda-cyhalothrin (Figure 4 (a) and (b)), although the others were not totally absent. Investigations by Mawussi (2008) also revealed the presence of organochlorine pesticides with concentrations lower than those in our study [21].

For other hand, samples from the Ogou River (M2a, M2b) and samples from the Anié River (M4a, M4b) were characterized with pesticides level above the standard (0.50 $\mu g.L^{-1}$) at the entrance to the study area. Nevertheless, the pesticides content increases twice for the sample located in the study area M2b in comparison to sample M2a which is at the entrance (*Figure 3*). The M4 samples contain a wide range of pesticides (50 to 80% of studied pesticides), unlike the M2 samples (< 50%) shown in *Figure 4(a)*. Samples M2a et M2b are dominated by chlorpyrifos at 70% of their total concentration. This can be explained by the agricultural practices in this area, which is dominated by soya mainly exported which does not require a wide variety of pesticides.

Rivers M3, M5, M6 and M7 all have their sources in the study area (*Figure 1*). Pesticides concentrations are all below the standards except for sample M5b (*Figure 3*) located at the junction of several streams. M5b were sampled in the river which flows through cotton and bean growing areas. These crops are usually treated mostly by insecticides more than five time per season. In overall, samples M3, M5, M6 and M7 were mostly concentrated in chlorpyrifos and lambda-cyhalothrin. However, the sample M3 has shown a strong content of deltamethrin.

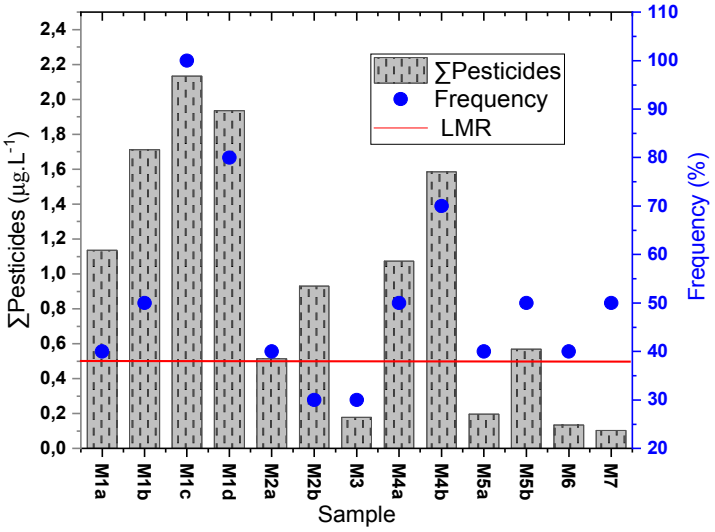
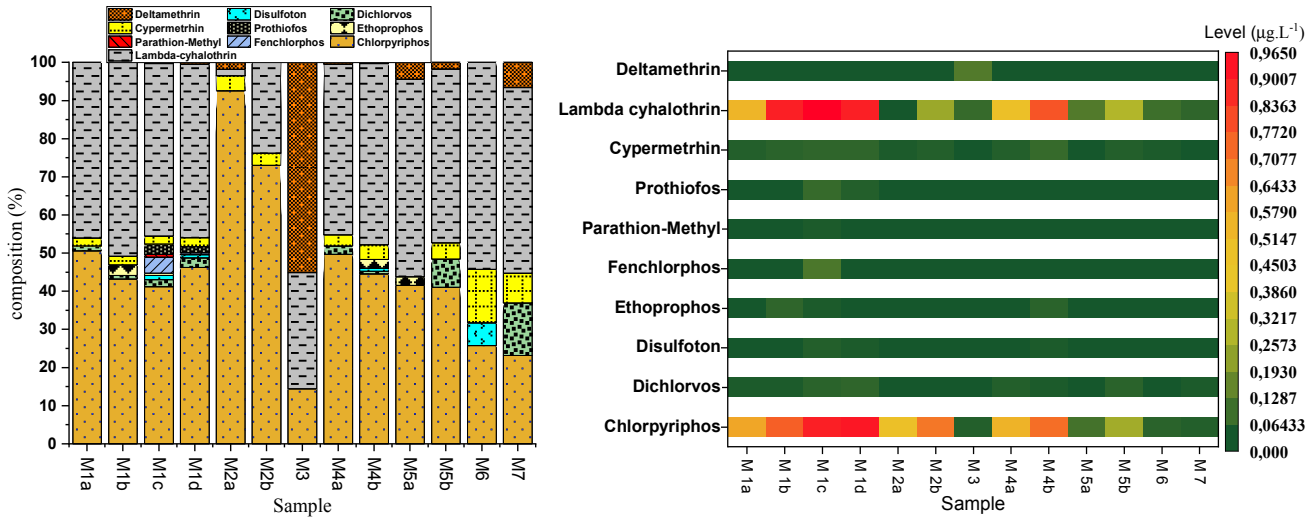


Figure 3. *Pesticide contamination of samples.*



(a) (b)

Figure 4. *Proportion of pesticide in each sample.*

3.4. Ecotoxicological Risk

The RQ values obtained for fish and crustaceans are shown in *Figure 5*. Fish RQs ranged from 0.0005 to 0.01305 corresponding to negligible risk. Only samples M1(b, c) and M2b showed low risk levels for fish. Previous studies carried out by Gbénonchi [22], have shown the presence of several organochlorines in fish from the Nangbeto dam to be above the maximum pesticide residue limit allowed by the WHO.

The risk for crustaceans ranged from 0.02028 to 0.3477. Samples (M1) and (M4) were characterized by a moderate level of risk for crustaceans. The frequency consumption of crustaceans from this area may cause long-term public health concerns.

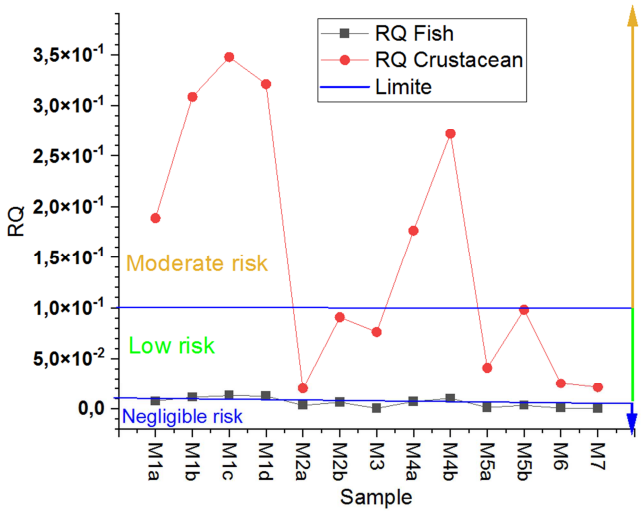


Figure 5. *Risk quotient for fish and crustaceans.*

4. Conclusion

The contamination of surface waters by several organophosphate and pyrethroid pesticides and the risk to fish and crustaceans were assessed in the middle part of the Mono river basin. The study revealed that all river waters in the basin were polluted by pesticides, mainly chlorpyrifos and lambda-cyhalothrin, highly available on markets. The agricultural practice in the area contributes enormous of the pesticides content in water samples. The risk to fish is negligible for all the analysed samples, while the risk to crustaceans is moderate for most of the larger rivers and low for the smaller rivers. Using these waters as untreated drinking water should be avoided, especially during the rainy season, because of their pesticide pollution levels. Farmers must be raised awareness for rational use of pesticides to reduce pollution.

Acknowledgments

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Conflicts of Interest

The authors declare no conflicts of interest.

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