Biodegradation of chlorpyrifos by autochthonous microbiota from a drinking water reservoir

Abstract

Chlorpyrifos, a pesticide widely applied in agricultural activities, may be transported from soil to surface water and even to groundwater by leaching and runoff due to physicochemical interactions between soil and this substance. Once in the environmental compartments, chlorpyrifos may be transformed by biotic and abiotic pathway reactions to increase the complex and multiplex effects over biota. Thus, biodegradation is an important natural reaction to reduce the environmental levels of pesticides and autochthonous microbiota with a high potential for the biotransformation of substances and could be related to the persistence and dynamics of chlorpyrifos. In this study, the biodegradation of chlorpyrifos under autochthonous microbiota in natural water from a reservoir was assayed. In samples, bacterial indicators such as Pseudomonas aeruginosa, Escherichia coli, and total coliforms were detected using selective culture medium and differential culture medium, such as Pseudalert and Colilert. Under aqueous medium with autochthonous microbiota, chlorpyrifos was transformed to 3,5,6-trichloro-2-pyridinol (TCP), and half-life was estimated at 10.2 h. Results showed that these bacterial indicators in the reservoir may reduce the chlorpyrifos levels in the water body but whether the metabolite TCP emerges in the water body with aquatic effects remains unknown.
Keywords: biodegradation; chlorpyrifos; pseudalert, colilert; autochthonous microbiota; surface water.

Biodegradación de clorpirifos por medio de microbiota autoctona de un embalse de agua potable

Resumen:

El clorpirifos, es un plaguicida aplicado ampliamente en actividades agrícolas. Esta sustancia, puede ser transportado desde el suelo a las aguas superficiales e incluso a las subterráneas por medio de lixiviación y escorrentía debido a las interacciones físicoquímicas suelo-sustancia. Una vez en los compartimentos ambientales, el clorpirifos puede transformarse mediante reacciones bióticas y abióticas, lo que aumenta los efectos complejos y múltiples sobre la biota. Así, la biodegradación es una reacción natural importante que reduce los niveles ambientales de plaguicidas y por lo tanto, la microbiota autóctona con alto potencial de biotransformación podría estar relacionada con la persistencia y dinámica del clorpirifos. En este estudio, se analizó la biodegradación de clorpirifos por medio de microbiota autóctona en agua natural de un embalse. En las muestras se detectaron indicadores bacterianos como Pseudomonas aeruginosa, Escherichia coli y coliformes totales utilizando medio de cultivo selectivo y medio de cultivo diferencial, tales como Pseudalert y Colilert. En medio acuoso con microbiota autóctona, el clorpirifos se transformó en 3,5,6-tricloro-2-piridinol (TCP), y la vida media se estimó en 10,2 h. Los resultados mostraron que estos indicadores bacterianos en el embalse pueden reducir los niveles de clorpirifos en el cuerpo de agua, pero aún se desconoce si el metabolito TCP emerge en el cuerpo de agua con efectos acuáticos.

Palabras claves: biodegradación; clorpirifos; pseudalert, colilert; microbiota autoctona; agua superficial.

1. Introduction

Pesticides are chemical substances widely applied in Colombian agriculture around many watersheds of important drinking water reservoirs. Some pesticides such as chlorpyrifos (CPF), diazinon, and malathion are organophosphorus pesticides (OPs) that are frequently
applied but not detected by conventional methods (Correa Zuluaga et al., 2018; Ramírez et al., 2023). However, using passive sampling methodologies such as semipermeable membrane devices (SPMDs) and the polar organic chemistry integrative sampler (POCIS), CPF has only been detected at levels closest to 5 ng L⁻¹, as well as the degradation product 3,5,6-trichloro-2-pyridinol (TCP), in Colombian reservoirs (Narvaez Valderrama et al., 2023). The degradation product TCP detected in the water body indicates that biotic and abiotic processes may induce but not completely degrade pesticides in reservoirs; therefore, the parent compounds cannot be detected (Narváez Valderrama et al., 2012). Therefore, the degradation products analysis should be the focus methodology for pesticide monitoring.

Some natural degradation processes, such as chemical hydrolysis and biodegradation, may be the most important processes to reduce pesticide levels but is accompanied by the emergence of degradation products (Bay Liu et al., 2001; Racke et al., 1996). For example, at similar pH conditions to that of surface water in reservoirs (pH = 10), CPF may be hydrolyzed to TCP with a half-life at 12.5 h (Narváez Valderrama et al., 2014)[7]. However, the complex influence of all-natural conditions may reduce the persistence of pesticides. Thus, the knowledge of radiation, pH and autochthonous microbiota may play an important role in degradation studies. In bacterial microbiota enzymatic reactions, enzymes such as hydrolases, esterases, phosphatases and dehalogenases, among others, participate in pesticide degradation (Kumar et al., 1996). In this aspect, in Colombian reservoirs, identified bacterial populations such as Staphylococcus spp and Bacillus spp related to Pseudomonas spp have been associated with pesticide biodegradation such as organochlorines and some organophosphates such as diazinon and methyl parathion (Cycoń et al., 2009; Nawab et al., 2003; RANI & LALITHAKUMAR, 2010). Therefore, in water bodies, it is possible to find high variability of heterotroph microbiota that may biodegrade pesticides. For example, the organophosphate CPF may be broken down by the phosphodiester bond, reducing it to TCP (Bootharaju & Pradeep, 2012). However, the final product from TCP could induce resistance in the biodegradation of high amounts of CPF because their metabolite may be more toxic for microbiota. Although the degradation of CPF reduces its cholinergic effects in acute toxicity,
the chronic and acute effects of TCP are incompletely understood. In this paper, autochthonous microbiota in reservoirs were studied by the detection and quantification methodologies of Pseudalert and Colilert under ISO/16266-1:2006 and ISO/9308-2:2012, respectively. The methodology applied allowed the growth of the microbiota population during the degradation time of CPF spiked in natural water from a reservoir. The data from this paper may be applied to identify the potential biodegradation of water matrices under the natural microbiota of pesticides and possible pathways for degradation.

2. Materials and methods

2.1. Chemical and materials

To detect and quantify Pseudomonas aeruginosa and Escherichia coli, we applied the Pseudalert and Colilert kits, respectively, which were purchased from IDEXX Laboratories and supplied by Aqualab Ltda-Santa Fe de Bogotá, Colombia. Additionally, the agar medium for total heterotroph analysis was supplied by Merck Millipore (Medellín, Colombia). For the analytical method, high-purity standards > 99.5% for CPF and TCP were purchased from Chem Service, Inc. (West Chester, PA, USA). Furthermore, pesticide analysis-grade solvents such as methanol (MeOH), acetonitrile, ethyl acetate and acetone were supplied by Burdick and Jackson (Honeywell) (Morristown, NJ, USA). Finally, derivatization was used for TCP analysis by gas chromatography–mass spectrometry (GC/MS) using N-(tert-butyldimethylsilyl)-N-methyl-trifluoroacetamide (MTBSTFA CAS 77377-52-7; Sigma-Aldrich, St. Louis, MO, USA). For TCP and CPF solid-phase extraction (SPE) from water, the cartridge Isolute 101 (2 mL) was used (Agilent Technologies, Santa Clara, CA, USA). All the standards were prepared in acetone at 1,000 mg L\(^{-1}\) and were stored at -20 °C.

2.2. Sampling and sample extraction

To study the potential biodegradation, an important tributary for a reservoir was sampled using a Kemmerer bottle at a depth close to 50
cm that was then stored in a glass amber bottle previously sterilized and kept at 8 °C.

For biodegradation analysis, 1 liter of natural water per duplicate was spiked with CPF to reach a final concentration of 1000 µg. L⁻¹ and this was labeled as spiked natural water (SNW). Additionally, 1 liter of natural water per duplicate was previously deactivated by autoclave to remove autochthonous microbiota and the sample was spiked with CPF at a similar level as that of SNW. The sample was labeled as deactivated microbiota by autoclave (DMA). Furthermore, 1 liter of natural water per duplicate was kept as a blank and the sample was labeled as non-spiked natural water (NSNW). All the samples were stored in the dark to avoid photolytic effects. Thereafter, 10 mL of water was extracted from the samples at 0, 3, 6, 9, 15, 24, 48, and 178 h for microbiology analysis, and an additional 10 mL was extracted at 0, 3, 6, 9, 15, 24, 48, 96, 144 and 178 h for GC/MS analysis of CPF.

2.3. Gas chromatography analysis

Ten milliliters of water sample extracted from the different treatments were passed through the SPE cartridge Isolute 101 that was previously conditioned with HCl (0.1 M): H2O: MeOH at a ratio of 2:2:2. Thereafter, the analytes were eluted using a mixture of ethyl acetate: acetone: acetonitrile at a ratio of 5:15:10. The collected fraction was concentrated by an evaporator, and the final volume was transferred into vials to be dried by nitrogen before injection. Finally, 1 µL of sample was analyzed by the GC/MS Thermo Scientific system (single quadrupole) by electron impact in the selected ion monitoring (SIM) mode and SCAN mode. For more details, the GC/MS conditions can be found in supplementary materials (table SM1).

For TCP analysis, the sample was derivatized by MTBSTFA according to the modified methodology (Holger et al., 2001). More details are presented in supplementary materials (figure SM1).
2.4. Microbiology analysis of autochthonous microbiota

For all microbiology analysis, 10 mL of previously extracted sample was used. Therefore, bacterial populations of *P. aeruginosa*, total coliforms, *E. coli* and total heterotrophs were assayed from samples SNW, NSNW and even blank samples with deactivated microbiota by autoclave (BDMA) using standard methods such as Colilert and Pseudalert under International ISO/9308-2:2012 and ISO/16266-1:2006, respectively. Additionally, a standard method to examine water and wastewater SM-9215-C was applied for total heterotroph analysis.

2.5. Data analysis

Analysis of all results was performed using the free software R, version 3.0.1 from 2013. Additionally, the software STATGRAPHICS Centurion XVI, version 17.1.17 (32 bits), and Microsoft Excel 2010 were used.

All models, including calibration curves and degradation kinetics, to quantify the goodness of fit were analyzed by the percent residual accuracy (%RA). Additionally, differences between the degradation treatments and blank were established by the relative standard deviation (%RSD). Finally, the half-life was estimated from a two-phase decay model because the outcome measured was the result of the sum of a fast and slow exponential decay. Therefore, the mathematical analysis is presented as follows (Equation 1):

\[
C_f = C_{i(fast)} \cdot e^{-k(fast)t} + C_{i(slow)} \cdot e^{-k(slow)t} + \text{plateau}
\]

Where \(C_f\) is the final concentration, \(C_i\) is the initial concentration at the fast time \((C_{i(fast)})\) or slow time \((C_{i(slow)})\), and \(k\) is the kinetic rate constant at the fast time \((k_{(fast)})\) or slow time \((k_{(slow)})\) in a time \(t\). Finally, the plateau is the Y value at infinite times, expressed in the same units as Y. According to the equation, two half-lives could be found but the values estimated a slow degradation for the latest times.
3. Results and discussion

3.1. Mass chromatography for quantification and identification

The separation method for CPF and TCP showed good selectivity for those target compounds. The mass ions applied in SIM and retention times ($t_R$) are presented in supplementary materials (table SM2). Additionally, for qualification, the mass spectrum was analyzed for levels higher than 30 µg. L$^{-1}$ to identify CPF biodegradation to TCP degradation. For more details, time retention ($t_R$), quantification ion, and detection ions can be found in Figure 1.

![Figure 1. Chromatography separation methods (GC/MS full SCAN) for CPF and TCP. The results presented were obtained for time zero in the biodegradation study of CPF spiked in samples. The identification by mass spectrometry showed the presence of target analytes in the extracted samples.](chart.png)

The quantification methods showed a limit of detection (LD) of 2.2 µg. L$^{-1}$ and 0.2 µg. L$^{-1}$ for CPF and TCP, respectively, while the limits of quantification (LQs) were estimated at 5.8 and 0.7 µg. L$^{-1}$ for the same substances, respectively. The values calculated in this paper for TCP were found to be four times higher than those values reported by Holger and coworkers at 0.05 µg. L$^{-1}$ for LD and 0.1 µg. L$^{-1}$ for LQ, indicating the sensitive and reproducible application of this methodology in the literature (Holger et al., 2001). Additionally, curve calibration presented $r^2 > 0.99$, indicating linearity for quantification for both analytes. Finally, the SPE was found to be accurate and was
calculated as a recovery percentage at three levels of extraction. The results are presented in Table 1.

### Table 1. Recovery study at different levels for target analytes

<table>
<thead>
<tr>
<th>Recovery for CPF</th>
<th>Recovery for TCP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Range</td>
<td>low</td>
</tr>
<tr>
<td>%RSD</td>
<td>7.5</td>
</tr>
<tr>
<td>Recovery</td>
<td>70.8</td>
</tr>
</tbody>
</table>

The values found for recovery extractions were found between 70 and 120%, including % RSD < 15% in trace analysis. Thus, the results of the recovery of TCP and CPF were found to be a reproducible and accurate extraction method. More details are presented in supplementary materials (Figure SM1 and figure SM2).

### 3.2. Standardization of microbiota analysis

The methods for Colilert and Pseudalert analyses were previously checked in the laboratory applying the normalized methods ISO 9308-2:2012 and ISO 16266-1:2006, respectively. All the checked parameters showed good repeatability, reproducibility and accuracy in methodology. The results for the acceptance criteria are presented in Table 2.

### Table 2. Results for the checked parameters in the Colilert methodology

<table>
<thead>
<tr>
<th>Checked parameter for Colilert method</th>
<th>Results</th>
<th>Acceptance criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Repeatability</td>
<td>Total coliforms</td>
<td>Total coliforms</td>
</tr>
<tr>
<td></td>
<td>&lt; 6%</td>
<td>%RSD &lt; 20%</td>
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<tr>
<td></td>
<td>Escherichia coli</td>
<td>Escherichia coli</td>
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<tr>
<td></td>
<td>&lt; 7%</td>
<td>%RSD &lt; 20%</td>
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</tbody>
</table>
### Checked parameter for Colilert method

<table>
<thead>
<tr>
<th>Intermediate Precision</th>
<th>Results</th>
<th>Acceptance criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total coliforms &lt; 6%</td>
<td></td>
<td>Total coliforms %RSD &lt; 20%</td>
</tr>
<tr>
<td><em>Escherichia coli</em></td>
<td>&lt; 6%</td>
<td><em>Escherichia coli</em> %RSD &lt; 20%</td>
</tr>
<tr>
<td>Accuracy</td>
<td>Total coliforms 98 &gt; % &lt; 128</td>
<td>Recovery percentage between 80%–120% for total coliforms and <em>Escherichia coli</em></td>
</tr>
<tr>
<td>Total coliforms</td>
<td>99 &gt; % &lt; 106</td>
<td></td>
</tr>
</tbody>
</table>

Similarly, all the results obtained in the checked parameter for the Pseudalert method showed good repeatability and reproducibility of the analyzed microbiota methods because the %RSD values found were < 4% for both parameters. Therefore, all the methodologies for microbiota analysis during the biodegradation study were considered reliable testing.

### 3.3. Biodegradation analysis of CPF

The total coliform analysis showed exponential growth until 50 h. At this time, the maximum percentage of colony-forming units (CFU) was found in non-spiked natural water (NSNW) and in spiked natural water with CPF (SNW) (Figure 2a and 2b). However, a higher CFU was found in the NSNW sample than in the SNW sample.

**Figure 2.** Exponential population growth study of autochthonous microbiota. A. Population growth study of persistent microbiota in NSNW. B. Population growth study of persistent microbiota in SNW. The percentages were estimated by the ratios between individual data estimated at different times and total CFU found in the assay.
The results for NSNW allowed estimation of the autochthonous microbiota in samples from the reservoir; thus, the bacterial population was possibly related to bio-degradation. During the population growth study, the CFU of E. coli was undetected by Colilert analysis but the CFU of P. aeruginosa was found in both treatments (NSNW and SNW) by Pseudalert analysis. More detail for detection in supplementary materials (Figure SM3 and SM4). The % CFU was decreased completely in NSNW at 178 h but not in SNW according to figure 2.

The analysis of heterotroph microbiota showed an exponential growth population until 50 h; however, thereafter, the population decreased to zero at 178 h in SNW and the heterotrophs stayed stable with a population higher than 6,500 CFU in NSNW (Figure 2). The results showed that, in the SNW sample, some degradation products or CPF may affect the population growth in microbiota because the population decreased after 50 h compared with that in the NSNW sample. However, a low CFU in some populations of autochthonous microbiota was found in the degradation study (Figure 2).

Finally, in the BDMA, microbiota was not detected, indicating the efficiency of the removal of bacterial populations. Thus, the autoclave was found to be appropriate as a non-biodegradation control during this study.

3.4. Chlorpyrifos and their metabolite analysis

In sample analysis of NSNW, CPF was not detected at an LD to 2.2 µg L⁻¹. Thus, the spiked fraction of this pesticide is the only source in the methodology applied. According to our results, the CPF was degraded by a first-kinetics model in SNW and even in the BDMA control. The graph analysis is presented in Figure 3.
The degradation analysis of CPF in SNW showed a significant difference (p value < 0.05) with BDMA samples regarding the higher rate degradation of CPF exposed to microbiota. For the biodegradation study, the dissolved oxygen concentration (DOC) was measured during time analysis, which decreased to reach 5.2 mg L⁻¹. Therefore, the degradation was performed under aerobic conditions. On the other hand, the pH values were also followed during the time study and were, on average, close to 8.2 units. This value was kept stable in analysis. Thus, the degradation presented in BDMA may be related to the hydrolysis of CPF and not the photodegradation effect because all samples were carried out under dark conditions. The parameters measured in all experiments, such as pH and DOC, are presented in Figure 4.
The half-life of CPF was 17.6 h in SNW but was 18.3 h in BDMA. Additionally, the percentage of CPF degraded was > 40% and the CPF was found to be degraded at values less than 25%. The results may be compared in Figure 3.

The metabolite TCP was detected in the biodegradation samples of SNW and was identified by mass spectroscopy at a $t_R$ of 8.08; thus, CPF was partially degraded in samples (Figure 1). However, in BDMA samples, the same degradation product was detected; therefore, hydrolysis may induce similar degradation pathways. The kinetic degradation is presented at short times in Figure 5.
Additionally, TCP was found at 43.5 µg. L⁻¹ in NSNW, which was considered a control sample for microbiota growth analysis. Thus, this sample showed the presence of this degradation product in natural water in reservoirs but not CPF. Therefore, the parent compound, according to our hypothesis, is effectively degraded under natural conditions and analytical methods should focus on degradation products monitoring. However, TCP is also degraded according to the first-kinetics order due to hydrolysis and biodegradation effects by autochthonous microbiota (Figure 6).

The kinetic degradation of TCP was applied to estimate the degradation constant \(k\) that was presented in Figure 6. Similarly, applying equation 1, the half-life (slow) was 10.2 h under natural conditions under dark conditions. By contrast, in SNW and BDMA, the percentage of TCP increased to reach the level at which this metabolite started to decrease (Figure 5).

After 24 h, the concentration of TCP reached the maximum value at 392 µg. L⁻¹ in the SNW sample and 98 µg. L⁻¹ in the BDMA sample.

Higher amounts of TCP found in SNW may be related to faster degradation of CPF under microbiota conditions and biodegradation processes. However, the decreasing levels followed first-order degradation in SNW, allowing the estimation of \(k\) and half-life presented in Figure 6.
The degradation time of TCP in SNW was faster than that in BDMA. Therefore, autochthonous microbiota may play an important role in TCP degradation after the partial degradation of CPF in natural waters. For kinetic degradation of TCP see the figure 7

![Figure 7. Formation and TCP degradation. A. TCP level variability at different times of analysis in SNW. B. TCP level variability at different times of analysis in BDMA. $k = 0.063$; half-life = 11.4 h](image)

4. Discussion

Many pesticides may be transferred to water bodies from soil or by direct discharge. However, many natural processes may degrade them to complex and multiplex degradation products (Fenner et al., 2013; Narváez Valderrama et al., 2012). Thus, dynamic studies for the risk assessment of parent compounds, such as studies of the bioaccumulation and environment levels, should focus on analyzing degradation products (Corellas et al., 2015; Liu et al., 2016). Knowledge about principal natural degradation factors may focus on methodologies to detect photoproducts by sunlight effects, hydrolytic products by pH effects, or metabolites by biodegradation. In this study, we investigated the biodegradation of CPF by autochthonous microbiota from a reservoir that could partially or completely degrade this pesticide. According to our results, natural water collected from some drinking water reservoirs contain bacterial populations, such as P. aeruginosa and E. coli, among other heterotrophs, that degrade CPF to the intermediate metabolite TCP. In this aspect, phosphotriesterase
enzymes purified and characterized from *P. diminuta* can hydrolyze organophosphate pesticides, such as diazinon and ethyl parathion (Caldwell & Raushel, 1991; Dumas et al., 1989; Munnecke, 1979). The results found in this comparative study between SNW and DMA show that although degradation of CPF occurred in both treatments, more pesticide was removed by biological processes. Thus, autochthonous microbiota in reservoirs play an important role in the environmental transformation fate of pesticides (Arbeli & Fuentes, 2007). However, the TCP arising in DMA indicates that hydrolytic processes occur at the same time by pH influence. In the recent literature, we found that CPF may be degraded at pH values higher than 7.8 with a half-life of 49 h (Narváez Valderrama et al., 2014). Therefore, the pH values were followed during the experiment but were not neutralized to maintain natural conditions due to multiplex factors that may affect pesticides in water bodies at the same time. The possible biochemical and hydrolysis events occurring in CPF upon biodegradation (under autochthonous bacteria) in water and hydrolytic medium is presented in Figure 8.

![Figure 8. Possible biodegradation pathway of CPF by biodegradation and hydrolytic effects.](image)

The autochthonous microbiota showed exponential growth in all treatments, but the bacterial populations started to decrease in SNW
after 24 h when the TCP started to be degraded. According to our results, TCP is a not toxic degradation product for autochthonous bacteria, a finding that was confirmed by NSNW treatment analysis because the bacterial populations continued growing despite the basal levels of TCP in the medium. Therefore, emerging unidentified degradation products may be associated with the decrease in the microbiota population. After photocatalytic treatment of pollutants, many degradation products may still induce endocrine disruptor effects an; thus, degradation does not imply the reduction of environmental risk (Ríos-Sossa et al., 2022)(See Figure 9 for the correlation study between the degradation of TCP and population growth).

The Pearson correlation study between the degradation of TCP and different autochthonous microbiota growth showed no differences between the groups (p value >0.05) in SNW treatment. Thus, bacteria may degrade CPF to reach the maximum level of their metabolite TCP; however, after TCP starts to be degraded, the population decreases. Additionally, during TCP formation, another sub-product, such as diethyl thiophosphate (a hydrolytic product), may be used as a phosphate source for microbiota (Figure 7). However, more unidentified metabolites in a complex mixture affect the autochthonous
microbiota after 24 h, as shown by the population decrease in Figure 2b. At the same time, TCP started to be biodegraded likely producing more toxic degradation products. For example, TCP may be degraded by the bacterial strain *Cupriavidus spp* to 2-pyridinol by dechlorination pathways with a possible source of the pyridine ring (Lu et al., 2013).

The half-life found for CPF was almost three times smaller than that estimated for hydrolytic condition testing in distilled water in a recent study conducted at a pH higher than 8 units [7]. Thus, the degradation of CPF under natural conditions may be the result of hydrolysis, biodegradation and even photo degradation. Although our experiments were performed under dark conditions, pH and microbiota could synergistically work. The results may explain why CPF is not easily detected in reservoirs in Colombia despite the widespread use in the watershed; thus, autochthonous bacteria in water bodies are efficient to transform CPF to TCP, which should be the marker in the quality water analysis of pesticides. Some reports showed that diazinon (organo-phosphorus pesticide) is degraded to 2-isopropyl-6-methylpyrimidine-4-ol (IMP); similarly, this substance should be the trace marker due to parent compound being undetected in reservoirs (Wang et al., 2019; Zhang et al., 2011). In this paper, the levels found in the sample taken from a natural drinking water reservoir was 43.5 µg L⁻¹ (data from the NSNW sample), which is related to our hypothesis about CPF degrading under natural conditions in reservoirs. Therefore, OPs may be transformed in reservoirs and studies about the risk assessment for complex degraded fractions of pesticides over aquatic species and even endocrine disruptor effects should be performed in future studies. Some pollutants may affect reproductive hormones even at low levels (Narvaez Valderrama et al., 2022)

5. Conclusions

Autochthonous microbiota, such as *P. aeruginosa*, Heterotroph, and *E. coli*, among other coliforms, were identified in the water bodies of reservoirs in Colombia and were found to degrade CPF by phosphodiester bond hydrolysis to TCP. Although this metabolite was not toxic for microbiota, after TCP degradation, many populations
were decreased possibly more via toxic and complex fractions of degradation products emerging. Thus, analytical methods and even risk assessment should be focused on degradation products and not CPF detection. The half-time of CPF and its metabolite TCP showed values of <12 h. Therefore, the reservoir may play an important role as a natural water bioreactor with hydrolytic and photolytic effects. Additionally, the complex toxicity of the degraded complex fraction at low levels of pesticides should be studied in future research because many aquatic species and even humans may be exposed.

6. Acknowledgments

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7. Conflict of interests

The authors declare that there is no conflict of interest with regard to the published results.

8. References


