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## Acute Toxicity of Chlorpyrifos on the Freshwater Bivalves (*Unio Tigridis*) and Effects on Bioindicators

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### Abstract:

A freshwater bivalve plays a crucial function in aquatic habitats as the filtered water and burrowing mussels mix the sediment, thus increasing oxygen content and making the ecosystem healthier. The aim of the study is to see how chlorpyrifos affects biochemical markers in freshwater mussel *Unio tigridis*. About 180 individuals per taxon and water samples were collected from the Qandil water resource on the Greater Zab River, Erbil Province, Iraq. Once arrived at the lab, the individuals were kept in aquaria with river water and an air-conditioned room Temperature: 25±2 and Light: 12h/12h and acclimatized to laboratory conditions for seven days in aged tap water. The mussel's identification molecularly and the DNA sequence of the mussel includes *U. tigridis* supplied gene bank accession number ON872361, ON872362, ON872363, and ON872364 nucleotide sequencing. The 96-h toxicity of chlorpyrifos pesticide in the freshwater mussel *U. tigridis* was investigated using various nominal concentrations, including 50, 100, 200, 300 and 400 ppm. The water quality of the river and aquaria was tested for physicochemical parameters including water temperature, the potential of hydrogen ion pH, electrical conductivity EC, and total dissolved solids TDS, dissolved oxygen, total alkalinity, total hardness, calcium ion, magnesium ion. Water quality results of aquaria revealed that most tested variables were favorable for the breeding of mussels. The mortality of the mussels was observed daily and the 96 h LC<sub>50</sub> value for mussels was 157.99 ppm. Within the tissue of the gills, Acetylcholinesterase (AChE), Glutathione S-transferase (GST), Catalase (CAT), and Malondialdehyde (MDA) were determined. The chlorpyrifos exposures caused significant increases in GST, CAT, and MDA. The elevation of oxidative stress biomarkers was inversely related to the AChE inhibition in the examined species. In conclusion water pollution by chlorpyrifos lead to unsafe condition for aquatic taxa.

**Keywords:** biochemicals, LC<sub>50</sub>, mussel, pesticide, toxic effect.

### Introduction:

Bivalves are a significant animal that has made excellent bioindicators for evaluating water pollution and figuring out its level in bodies of water<sup>1</sup>. Stormwater runoff, untreated wastewater discharges, agriculture, and air deposition are just a few of the various ways that pollutants infiltrate water systems. Around the world, non-point source nutrient enrichment has taken over as the main cause of deteriorating water quality<sup>2</sup>. Pesticides and heavy metals are frequent freshwater pollutants that have a direct impact on aquatic life. Pesticides have a variety of negative consequences on public health, the ecosystem, the quality of food, and biodiversity. Pesticide poisoning of the environment ranks among the most significant issues facing the country due to its high persistence, potential for extreme toxicity,

and slow breakdown. Pesticide employment is growing in farming, which causes environmental pollution. The majority of pesticides cause oxidative stress by disrupting the body's normal antioxidant system<sup>3</sup>. The sensitivity of freshwater species to organophosphates pesticides varies widely according to chemical compositions, duration of exposure, quality of water and taxa. In general, pesticide adversely affects non-target species, such as benthic invertebrates and fish, that live in the freshwater ecosystem due to their watery dispersal<sup>4</sup>. Pesticides are routinely identified in the aquatic ecosystem as artificial toxins; organophosphates and carbamates are new chemically synthesized insecticides that are strong neurotoxic chemicals. Chlorpyrifos is a common ingredient in organophosphates pesticides, and it's utilized on a

vast range of products including wheat, fruit and vegetables<sup>5</sup>. Pesticide overuse has a variety of negative consequences on the environment and the creatures that are exposed to it, which has garnered interest in scientific fields. The inadequacy of a study based on empirical data on the opinions of farmers is one of Erbil's biggest issues. Typically, farmers are not well informed about the procedures for choosing and applying pesticides<sup>6</sup>.

Bivalves are generally long-lived between 15 to 40 years and have comparatively low mobility in their natural habitat. These creatures filter-feed on minute fine particles in the aquatic environment, but they have additionally been observed depositing food in the benthic habitats<sup>7</sup>. Mollusca, specifically bivalves, has played a significant influence in determining the rates of pollutants around the planet. This is due to the tactical advantages of collecting, global dispersion, generally quiescent behaviors, appropriate size, and, in many cases, environmental and socioeconomic significance<sup>8</sup>. A biomarker is a fundamental change that may be detected and/or assessed by bivalves at the genetic, metabolic, cell, physiological, or behavioral level and that discloses an organism present or the previous response to at least a single chemical in the ecosystem<sup>9</sup>.

Antioxidant enzymes are among the most popular markers utilized in pollution level monitoring. Among the first chemicals to also be utilized as a lipid peroxidation indicator was malondialdehyde (MDA)<sup>10</sup>. One of the important steps in oxidative stress is the modification of membrane phospholipids by lipid peroxidation, which is one of the indicators of stress. Chemically produced toxicity is frequently characterized by oxidative stress<sup>11</sup>. An enzyme called acetylcholinesterase (AChE) hydrolyzes acetylcholine and turns it into choline and acetic acid<sup>12</sup>. The enzyme regulates ionic currents in excitable membranes and is crucial for nerve conduction at the neuromuscular junction. The mechanism of the harmful effect of organophosphate insecticides is closely linked to the suppression of AChE. AChE has been and continues to be a widely utilized biomarker in freshwater ecotoxicity investigations, and it may also be employed as a neurotoxicity biomarker in species<sup>10</sup>. Glutathione S-transferase (GST) are detoxification enzymes that catalyze the attachment of glutathione (GSH) to a range of electrophilic molecules for the removal of potentially harmful xenobiotics. GST is found in the cytosol and microsomal fractions of cells<sup>13</sup>. Antioxidant enzymes like catalase (CAT) are part of the antioxidant defense system. By eliminating reactive oxygen species, this antioxidant enzyme helps to

maintain cellular homeostasis and antioxidant defense<sup>14</sup>. The purpose of this research is to see how chlorpyrifos affects biochemical markers in freshwater mussel *Unio tigridis* gill tissue.

## Materials and Methods:

### Water Quality

In September 2021, polyethylene bottles were used for the collection of river water at the study location. Water samples were examined for physical-chemical characteristics instantly such as water temperature °C, the potential of hydrogen ion pH, electrical conductivity EC ( $\mu\text{S}/\text{cm}$ ), and total dissolved solids TDS (mg/l) at the sampling site. Dissolved oxygen (mg/l), total alkalinity (mg  $\text{CaCO}_3/\text{l}$ ), total hardness (mg  $\text{CaCO}_3/\text{l}$ ), calcium ion (mg/l), magnesium ion (mg/l), were measured as soon as arrived at the laboratory according to the procedures of<sup>15,16</sup>. The following water parameters analyzed in each glass aquarium were pH, EC, TDS, DO, total alkalinity, total hardness,  $\text{Ca}^{2+}$ , and  $\text{Mg}^{2+}$  utilizing the techniques listed above. Throughout the test, every 48 h, all parameters were measured.

### Bivalve's Sampling and Acclimatization

The freshwater mussels *U. tigridis* were collected in September 2021 from Qandil water resources on the Greater Zab River, Erbil Province, Iraq  $36^{\circ}37'39.55''\text{ N } 44^{\circ}10'51.80''\text{ E}$ .

One species of freshwater mussel's hand-collected *U. tigridis* 7.5 cm long, 4.5 cm wide, and 26.2 g weight was found in the Greater Zab River's sediment area Fig. 1, branch in the Qandil area. About 180 individuals per taxon were sampled from the study area, cleaned, and transported to the laboratory with river water. Once arrived at the lab, the individuals were kept in aquaria with river water and an air-conditioned room Temperature:  $25\pm 2$  and Light: 12h/12h and acclimatized to laboratory conditions for seven days in aged tap water. During animal rearing time, the water was changed every 24 h. Mussels were not fed during the experiment period. The sample of mussels was identified using the common keys<sup>17,18</sup>.



Figure 1. *Unio tigridis* morphological view.

### Molecular Study

Four replications of a Bivalvia *U. tigridis* were employed for DNA sequencing after morphological identification. Genomic DNA from adductor muscle was isolated and purified using the GeneAll® Exgene™ for Clinic Cell SV small kit (Songpa-gu, Seoul, Korea). Agar gel electrophoresis was used to gauge the amount of genomic DNA that had been extracted before PCR<sup>19</sup>. The Nano Drop

1000 spectrophotometer is prepared to measure the optical density of a DNA sample as well as the concentration and purity of genomic DNA extraction to determine the amount and purity of DNA. The COI gene was successfully amplified using the primer pair LCO1490: 5'-GGTCAACAAATCATAAAGATATTGG-3' and HC02198: 5'-TAAACTTCAGGGTGACCAAAAAATCA-3', which were ordered from Macrogen (Korea) Table 1<sup>20</sup>. The PCR experiment was conducted in a 50 µL reaction cocktail including 25 µL of 2× master mix (AMPLIQON, Denmark), 1.0 µL of each primer 10 pmol, and 3 µL of genome template. The amount reached 50 µL using PCR-grade water. To verify that the DNA templates were completely denaturated, DNA amplification for the COI gene was carried out in the thermal cycler for 5 minutes at 94 °C. The PCR was then carried out as follows: 94°C for denaturation for 50 sec, 50°C for annealing for 45 sec, and 72°C for an extension for 50 sec. These parts were repeated forty times, with a 7-minute extension at 72°C as the final cycle. 0.8% of agarose gel electrophoresis in 1× TAE buffer was used to examine the PCR products<sup>21</sup>. and were sent to (Macrogen/South Korea) for sequencing. MEGA software was used to analyze and alter the sequences that were acquired.

Table 1. LCO1490 (forward) and HCO2198 (reverse) primers.

Direction	Name	Sequence
Forward	LCO1490	5'-GGTCAACAAATCATAAAGATATTGG-3'
Reverse	HC02198	5'-TAAACTTCAGGGTGACCAAAAAATCA-3'

### Experimental Design

The experiment consisted of 18 glass aquaria three controls and three replications of each other who received different exposure concentrations of chlorpyrifos 50, 100, 200, 300 and 400 mg/L. After applying chlorpyrifos immediately into the aquaria, they were left at 25 °C with a 12-h. darkness: 12 h. light cycle. The number of *U. tigridis* living and dead were recorded after 24, 48, 72, and 96 h by observing the mobility of individuals, the entry and escape of soft tissues from their shells in particular. The individual that couldn't enter their soft tissues and close their shells were considered as dead. Dead mussels were taken out of glass aquariums every day until the test was finished. Probit Analysis was used to measure the 96-h LC<sub>50</sub> of chlorpyrifos following<sup>22</sup>. Additionally, the mortality percentage was calculated as a following Eq. 1:

$$\text{Mortality \%} = \frac{\text{No. of death animals}}{\text{Total no. of animals at the beginning of the test}} \times 100$$

### Biochemical Analysis

Mussels were dissected, and the gills of each *U. tigridis* were cleaned in an ice-cold saline fluid. A handheld glass homogenizer was used to homogenize the samples in a phosphate buffer. The homogenates were centrifuged at 4,000 rpm for 10 min at 4 °C. Before testing, the supernatants were taken and stored at -80°C<sup>23</sup>.

For Acetylcholinesterase AChE: BC 2020 and Glutathione S Transfer GST: BC0350, concentrations were measured by colorimetric method using commercial assay kits according to the manufacturer's protocol of Beijing Solarbio Science and Technology Co., Ltd. But, the tissue Catalase (CAT) activity was measured using the technique of<sup>24</sup>. Finally, the lipid peroxidation level in the tissue

was quantified as malondialdehyde (MDA) according to the method of <sup>25</sup>.

### Data Analysis

Data were analyzed by using the SPSS program version 25. Analysis of variance (ANOVA) is one way used to handle water quality and biochemical data. To look for significant variations between the treatments, Duncan's post hoc test was used. For statistical significance, a p-value of 0.05 is used as the limit.

### Results and Discussion:

It appears vital to assess the harmful effects of pesticides on aquatic creatures like freshwater

mussels because there are numerous ways for environmental pollutants to infiltrate surface waters. The toxicity of the chlorpyrifos on *U. tigridis* biochemical markers is thus assessed in this work.

The mussel identification molecularly by utilizing comprehensive or particular initial gene magnification was uniform with phenotypic assessment. Data for the molecular sample was provided by CDS nucleotide sequencing, which also provided isolation diagnostic and minute properties. The DNA sequence of the mussel includes *U. tigridis* supplied gene bank accession number ON872361, ON872362, ON872363, and ON872364 nucleotide sequencing Fig. 2.

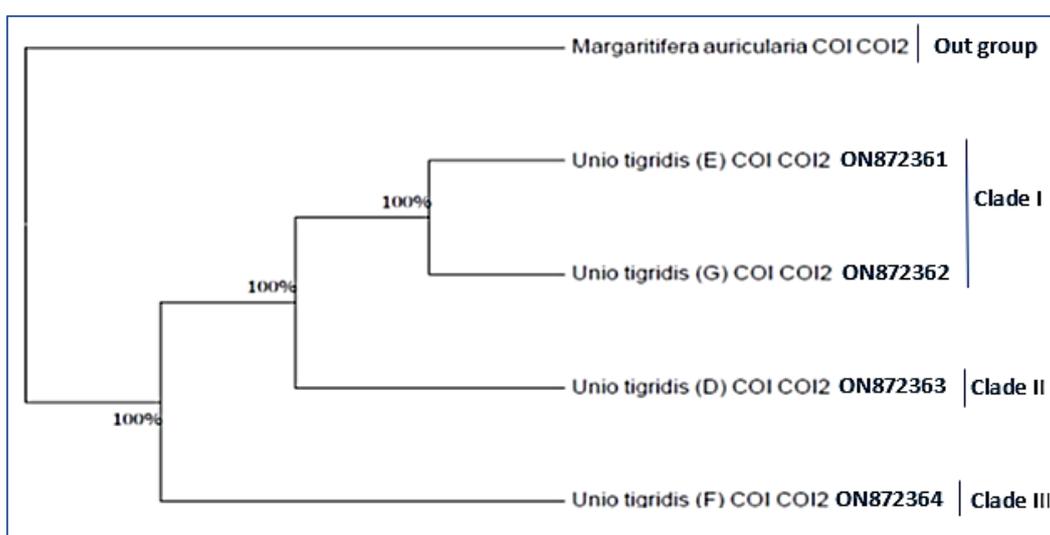


Figure 2. Analysis of COI's molecular phylogeny using the Maximum Likelihood approach.

The values of water quality parameters in Qandil water resources during the study are represented in Fig.3. Environmental elements like physical and chemical variables have an impact on the ecology of freshwater mussels <sup>26</sup>.

While Table 2, summarizes the value of aquaria water quality parameters. During a laboratory experiment, these parameters must fall

within the acceptable limits for aquatic species, especially for mussels' life. Water quality results of aquaria revealed that most tested variables were favorable for the breeding of mussels and were steady over the entire test. A control survival rate of 90% or more is typically required for short-term acute experiments with fish and invertebrates like bivalves <sup>16</sup>.

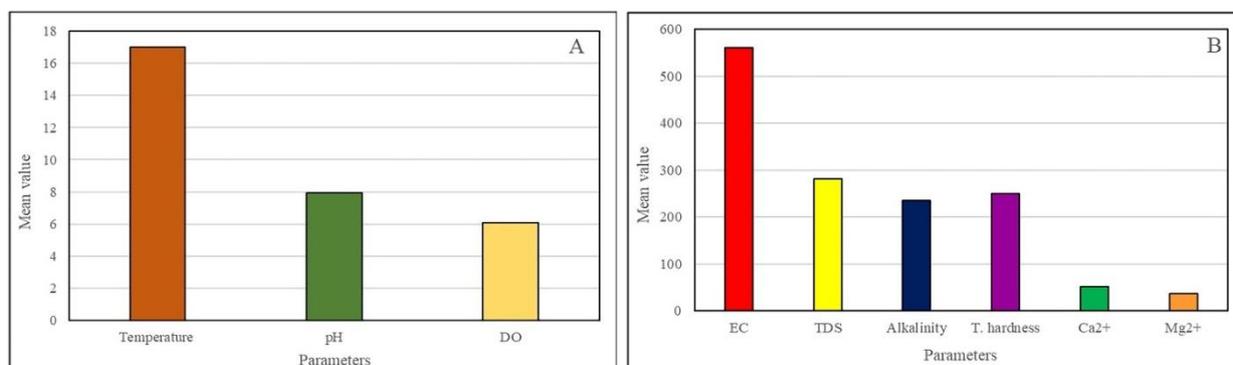


Figure 3. Mean values of physical and chemical parameters at Qandil water resource included A: Water temperature (°C), pH, DO (mg/l); B: EC (µS/cm), TDS (mg/l), Alkalinity (mg CaCO<sub>3</sub>/l), Total Hardness (mg CaCO<sub>3</sub>/l), Ca<sup>2+</sup> (mg/l) and Mg<sup>2+</sup> (mg/l).

**Table 2. Aquaria water quality parameters.**

Parameters	Range	Mean ± SD
pH	7.97-8.29	8.17±0.12
EC $\mu\text{S.cm}^{-1}$	503-561	546±21.73
TDS $\text{mg.l}^{-1}$	337-376	366±14.56
DO $\text{mg.l}^{-1}$	4.53-6.37	5.25±0.72
Alkalinity $\text{mg CaCO}_3.\text{l}^{-1}$	179-222	201±16.74
Hardness $\text{mg CaCO}_3.\text{l}^{-1}$	231-260	249±17.26
$\text{Ca}^{2+}$ $\text{mg.l}^{-1}$	65.17-79.07	73.40±5.24
$\text{Mg}^{2+}$ $\text{mg.l}^{-1}$	18.92-24.91	22.08±3.87

During the investigation, overproduction of mucus and a lengthening of shell closure was observed in all treated groups. These findings may help to explain why the investigated mussels had a defensive response, as other researchers found a similar pattern of behavior. Consequently, the findings of our investigation are consistent with <sup>27</sup>. As a typical response to stress, the mollusks secrete more mucus <sup>28</sup>. The most popular toxicity test is the 96 h LC50, which quantifies the toxicant level that,

after 96 h of exposure, results in a 50% lethal response <sup>29</sup>. This test was used in the current experiment, and from the results 96 h LC50 for *U. tigridis* treated with five concentrations of chlorpyrifos was 157.99 ppm. Numerous investigations have found 96-h LC50 values of chlorpyrifos for various freshwater mussel and other organisms, among them <sup>30-32</sup>. The result of the mortality percentage illustrated at Table 3.

**Table 3. Mortality percentage of *Unio tigridis*.**

Concentration (ppm)	24h	48h	72h	96h
0	0	0	0	0
50	0	10	20	20
100	10	10	20	30
200	0	40	60	80
300	0	40	70	80
400	20	50	80	100

The effect of chlorpyrifos on oxidative stress-related toxicity was studied including measurement of AChE, GST, CAT activities (U/mg protein), and MDA level (nmol/g protein) in gill tissue of freshwater mussels *U. tigridis*. Acetylcholinesterase (AChE) inhibition is the primary poisonous property of chlorpyrifos <sup>33</sup>. Table 4 shows how the organophosphorus pesticide chlorpyrifos affects AChE activity. The outcomes showed that the AChE activity in the gills of

pesticide-exposed mussels had significantly decreased  $p \leq 0.05$  with increasing concentration of pesticide in aquaria. The AChE activity declined gradually from 24 h to 96 h of exposure. Indeed, the least activities were observed after 96 h of exposure with a value of  $227.93 \pm 3.51$  U/mg protein compared to control with a value of  $492.81 \pm 0.54$  U/mg protein. The findings of the present study are consistent with an earlier study of the inhibitory effect of this type of pesticide on mussels' AChE <sup>34</sup>.

**Table 4. Evaluation of AChE (U/mg protein) (Mean ± SE) in *Unio tigridis* during different times in vivo exposed to different chlorpyrifos (CPF) concentrations.**

Concentration (ppm)	24 h	48 h	72 h	96 h
0	492.81±0.54 <sup>a</sup>	488.37±0.37 <sup>c</sup>	490.51±0.83 <sup>b</sup>	451.11±0.61 <sup>d</sup>
50	467.37±2.67 <sup>a</sup>	315.67±1.30 <sup>b</sup>	316.46±2.71 <sup>b</sup>	307.85±0.65 <sup>c</sup>
100	450.99±1.30 <sup>a</sup>	321.52±7.92 <sup>b</sup>	305.92±1.99 <sup>c</sup>	284.58±2.07 <sup>d</sup>
200	436.56±0.69 <sup>a</sup>	254.12±5.44 <sup>d</sup>	316.79±2.82 <sup>c</sup>	361.30±25.51 <sup>b</sup>
300	374.17±3.40 <sup>a</sup>	280.58±0.67 <sup>b</sup>	292.02±0.65 <sup>b</sup>	262.70±7.89 <sup>c</sup>
400	342.37±3.20 <sup>a</sup>	259.47±1.31 <sup>c</sup>	304.87±0.45 <sup>b</sup>	227.93±3.51 <sup>d</sup>

Note: Values in each row with different letters are significantly different, while values with the same letters are not significantly different.

Glutathione S-transferase (GST) activity assessed in control and treated gill *U. tigridis* throughout the experiment period is displayed in Table 5. Indeed, pesticides raised GST activity in gills to  $7.37 \pm 0.26$  U/mg protein compared to the control value of  $2.22 \pm 0.11$  U/mg protein. A significant rise  $p \leq 0.05$  was noticed in most

concentrations when compared with the gills of control mussels<sup>35</sup>. suggest that GST is crucial for detoxification and for preserving cells from oxidative damage. The detoxification of organophosphorus substances is facilitated by GST, which is essential for pesticide resistance. Similar results were observed by<sup>36</sup>.

**Table 5. Evaluation of GST (U/mg protein) (Mean  $\pm$  SE) in *Unio tigridis* during different times in vivo exposed to different chlorpyrifos (CPF) concentrations.**

Concentration (ppm)	24 h	48 h	72 h	96 h
0	$2.22 \pm 0.11^b$	$2.34 \pm 0.01^b$	$2.27 \pm 0.03^b$	$2.56 \pm 0.04^a$
50	$2.54 \pm 0.01^c$	$3.38 \pm 0.10^b$	$3.88 \pm 0.06^a$	$4.04 \pm 0.03^a$
100	$2.81 \pm 0.04^d$	$3.99 \pm 0.01^c$	$4.33 \pm 0.04^b$	$5.09 \pm 0^a$
200	$2.95 \pm 0.003^c$	$4.69 \pm 0.30^b$	$5.78 \pm 0.02^a$	$6.01 \pm 0.59^a$
300	$3.95 \pm 0.01^c$	$5.41 \pm 0.03^b$	$5.69 \pm 0.30^b$	$6.72 \pm 0.28^a$
400	$4.56 \pm 0.15^c$	$5.89 \pm 0.06^b$	$6.48 \pm 0.26^b$	$7.37 \pm 0.26^a$

Note: Values in each row with different letters are significantly different, while values with the same letters are not significantly different.

Additionally, variations in catalase (CAT) activity were seen as a result of chlorpyrifos exposure and its effects on the gills of mussels, *U. tigridis* Table 6. There is an obvious difference between the treatment and control groups. With an increase in chlorpyrifos dosage exposure, there were significant increases in CAT activity. At 400 ppm,

the maximum CAT activity 45.90 U/mg protein was observed. The tendency in CAT activity is consistent with earlier studies that found that certain insecticides increased CAT activity. This increase in CAT activity in the gill's aids in the pesticide's detoxification<sup>37</sup>. Our results come to agree with those of<sup>38(3)</sup>.

**Table 6. Evaluation of CAT (U/mg protein) (Mean  $\pm$  SE) in *Unio tigridis* during different times in vivo exposed to different chlorpyrifos (CPF) concentrations.**

Concentration (ppm)	24 h	48 h	72 h	96 h
0	$21.99 \pm 0.06^{ab}$	$21.56 \pm 0.39^b$	$21.06 \pm 0.50^b$	$22.81 \pm 0.24^a$
50	$32.62 \pm 0.50^b$	$34.22 \pm 0.17^a$	$32.51 \pm 0.26^b$	$34.73 \pm 0.32^a$
100	$35.36 \pm 1.10^c$	$36.48 \pm 0.41^{bc}$	$37.58 \pm 0.19^{ab}$	$38.98 \pm 0.06^a$
200	$37.00 \pm 1.00^b$	$39.02 \pm 0.07^b$	$38.93 \pm 0.07^b$	$42.37 \pm 1.23^a$
300	$39.97 \pm 0.04^b$	$35.97 \pm 0.54^c$	$37.48 \pm 0.26^c$	$43.70 \pm 1.35^a$
400	$42.74 \pm 0.34^b$	$42.45 \pm 0.87^b$	$40.51 \pm 0.30^c$	$45.90 \pm 0.47^a$

Note: Values in each row with different letters are significantly different, while values with the same letters are not significantly different.

Among the several aldehydes and ketones that are produced when monounsaturated and polyunsaturated fatty acids are per-oxidized is MDA. The maximum value of MDA  $1.64 \pm 0.01$  nmol/g protein was observed in an aquarium with a 400 ppm chlorpyrifos level during the 48 h of the test Table 7. A gradual increase in MDA levels was observed with increasing the concentration of pesticide during the

test period. The mechanism for the rise in MDA may reveal that insecticide may enter the cell's lipid membrane and disrupt the orientation of the phospholipids, altering the fluidity of the membrane. Lipid peroxidation rises as a result of oxidative damage that tissue or cell is unable to prevent, as indicated by a rise in MDA levels<sup>39</sup>. Our results come to agree with that of<sup>38</sup>.

**Table 7. Evaluation of MDA (nmol/g protein) (Mean ± SE) in *Unio tigridis* during different times in vivo exposed to different chlorpyrifos (CPF) concentrations.**

Concentration (ppm)	24 h	48 h	72 h	96 h
0	0.50±00	0.50±00	0.50±00	0.50±00
50	0.83±0.003 <sup>c</sup>	0.92±0.01 <sup>a</sup>	0.95±0.01 <sup>a</sup>	0.87±0.02 <sup>b</sup>
100	0.99±0.002 <sup>b</sup>	1.15±0.02 <sup>a</sup>	1.12±0.06 <sup>a</sup>	1.00±0.00 <sup>b</sup>
200	1.26±0.02 <sup>c</sup>	1.36±0.02 <sup>b</sup>	1.43±0.01 <sup>a</sup>	1.17±0.02 <sup>d</sup>
300	1.37±0.02 <sup>b</sup>	1.47±0.01 <sup>a</sup>	1.33±0.01 <sup>b</sup>	1.27±0.02 <sup>c</sup>
400	1.63±0.07 <sup>a</sup>	1.64±0.01 <sup>a</sup>	1.53±0.2 <sup>ab</sup>	1.40±0.04 <sup>b</sup>

Note: Values in each row with different letters are significantly different, while values with the same letters are not significantly different.

### Conclusion:

The organophosphorus pesticide chlorpyrifos is toxic to *U. tigridis* at different concentrations. The biochemical impacts of sub-lethal concentrations of up to 400 ppm of chlorpyrifos pesticides were evaluated in *U. tigridis* under laboratory conditions. The chlorpyrifos exposures caused significant increases in GST, CAT, and MDA. The elevation of oxidative stress biomarkers was inversely related to the AChE inhibition in the examined species. Our findings suggest the value of using biochemical and oxidative biomarkers to identify the harmful outcomes and toxicological processes brought on by environmental contaminants. Water pollution by chlorpyrifos from human activities, especially agriculture leads to unsafe conditions for aquatic taxa.

### Authors' declaration:

- Conflicts of Interest: None.
- We hereby confirm that all the Figures and Tables in the manuscript are ours. Besides, the Figures and images, which are not ours, have been given the permission for re-publication attached with the manuscript.
- Ethical Clearance: The project was approved by the local ethical committee in University of Salahaddin.

### Authors' contributions statement:

This work was carried out in collaboration between two authors. Y. A. Sh. designed the work, and N. S. H. collected the samples and did the experiment. Data analysis and the article were written by N. S. H. under the supervision of Y. A. Sh.

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## سمية الكلوربيريفوس على ذوات الصدفتين في المياه العذبة (*Unio tigris*) وتأثيراتها على المؤشرات الحيوية

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### الخلاصة:

للذوات الصدفتين وظيفة هامة في المياه العذبة إذ تقوم بعملية تصفية المياه ولكون مكوناتهم في البيئة القعرية يؤثر على زيادة نسبة الأوكسجين مما يؤدي الى بيئة قاعية اكثر حيوية. الهدف من الدراسة هو معرفة كيفية تأثير الكلوربيريفوس على الخصائص الكيميائية الحيوية في ذات الصدفتين في المياه العذبة *Unio tigris*. تم جمع حوالي 180 كائن إضافة إلى عينة من موارد مياه قنديل على نهر الزاب الكبير في محافظة اربيل - العراق. بمجرد وصولهم إلى المختبر ، تم الاحتفاظ بالأفراد في أحواض مع مياه النهر ودرجة حرارة الغرفة المكيفة: 25 ± 2 والضوء: 12 ساعة / 12 ساعة والتأقلم مع ظروف المختبر لمدة سبعة أيام في ماء منزوع الكلور. يتضمن تحديد بلح البحر جزئياً وتسلسل الحمض النووي لبلح البحر رقم انضمام بنك الجينات المقدم من *U. tigris* ON872361 و ON872362 و ON872363 و ON872364. تم فحص سمية مبيد كلوربيريفوس لمدة ستة و تسعون ساعة وتأثيرها على *U. Tigris* من خلال استخدام التراكيز المختلفة تتراوح ما بين 50, 100, 200, 300 and 400 جزء في المليون. تم تقييم الصفات الفيزيائية والكيميائية لمياه النهر والاحواض بما في ذلك درجة حرارة وإ الأس الهيدروجيني ، والتوصيل الكهربائي ، وإجمالي المواد الصلبة الذائبة ، والأوكسجين المذاب ، والقاعدية، و العسرة ، وأيون الكالسيوم ، وأيون المغنيسيوم. و أظهرت نتائج فحص مياه الأحواض بأنها مناسبة لتربية الكائنات، ثم تم ملاحظة الكائنات الميتة وكانت قيمة التركيز المميت النصفى بعد 96 ساعة لذات الصدفتين 157.99 جزء في المليون. وضمن خياشيم الكائن المدروس تم فحص AChE, GST, CAT, و MDA . وكانت النتيجة بأن تعرض للكلوربيريفوس يؤدي إلى الزيادة في GTS, CAT, MDA ولكن ارتبط ارتفاع مؤشر حيوية للإجهاد التأكسدي عكسياً مع AChE في الكائن المدروس. و تم استنتاج بأن تلوث المياه بالكلوربيريفوس يؤدي إلى ظروف غير آمنة للكائنات المائية.

الكلمات المفتاحية: الكيماويات الحيوية ، التركيز المميت النصفى ، محار ، مبيدات الآفات ، التأثير السام.