Study the effect of herbicide 2-4 D amine on a fish *Cyprinus carpio* Linnaeus (common carp)

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Abstract

This study was conducted to determine the impact of herbicide 2-4 D amine on a fish *Cyprinus carpio* Linnaeus (common carp), divided the carp into two categories depending on age, as it was over the age of the first category (45-90) days, and the category for a second was lifetime (90-150) days, were exposed to each category of fish to three concentrations of the pesticide which is (2%, 4% and 6%) and by three replicates for each concentration as well as a sample control, were also measured some properties physical and chemical water basins used in the experiment and these qualities are the water temperature, pH, electrical conductivity EC, dissolved oxygen, salinity and Hardness before and after the fish has also been used by the air pump is a device for generating oxygen within the basin.

The results of the current study, the presence of signs of superficial fish during exposure to a pesticide and in all concentrations and are these symptoms obtaining bleeding in Gills and the skin in addition to the presence of warp in the neck of the tail and the fall of the scales, but from a behavioral standpoint, it was noted irritability or confusion is clear for fish exposed to pesticides compared to a sample of control . Concentration was calculated as half the killer LC50 of the pesticide, which was (2.7938% and 1.5591%) for the two categories I and II, respectively.

The results of the study affected the first category of fish more than the second category and this is evident by the results of statistical analysis that showed a correlation inverse association between the toxicity of the pesticide and between the ages of fish used, As shown by the results of statistical analysis no significant differences at the level of probability (P <0.05) between the two categories of fish used in the experiment.

The results showed that the higher concentration of pesticide is higher the mortality rate and this is clear from the results of statistical analysis which showed a positive correlation between both categories and the concentration of the pesticide and the proportion of deaths (toxicity) .

*Keywords* / herbicide, 2-4 D amine, common carp, LC50 and the Physical Chemical characteristics.

Introduction:

Taking the use of herbicide enshrined increasing significantly resulting in the emergence of side effects represented in a breach occurrence in the normal balance for Ecosystems and that herbicides are a potential risk of a threat different forms of life including Human (Edward & Thompson , 1973)

The natural of water is Environmental center that receives large quantities of herbicides after their use for agricultural purposes and medical result of many natural processes like carrying air and washed from the soil by runoff water . It concentrations took of herbicides in natural aquatic environments are increasing cause was pollution of them (Solomon , et.al., 1996; Guiho, 1995; Oemichen & Habere, 1986) The result of high concentrations of herbicides occurrence of adverse effects on loved ones in different organisms in aquatic Ecosystems, resulting in a decrease in Communities of phytoplankton and zooplankton and fish (Dewey, 1986; Shafer , et
Protection of environment and water quality: the basis for agricultural production, Food Security and sustainable development 2012

In addition, causing of natural water pollution by herbicides lead to accumulation in the organisms tissues including fish to high levels pose a risk to human health when consumed (Osfor, et al., 1998).

The constant flow of agricultural effluent discharge into water bodies often lead to a variety of pollutant accumulation, which becomes apparent when considering toxic pollution (Mason, 1991). The application of environmental toxicology studies on non-mammalian vertebrates is rapidly expanding for the evaluation of the effects of noxious compounds. The indiscriminate use of herbicide, careless handing, accidental spillage, or discharges of treated effluents into natural waterways have harmful effects on the fish population and other forms of aquatic life and may contribute long term effects in the environment (Akhlar, 1986).

Herbicides are widely used for the control of water plants, which may impede the flow of water during the summer, when sudden heavy rain can flooding (Annune, et al., 1994) While the direct effect of herbicides addition is the loss of macrophytes, non-target organisms such as fish may also be affected through loss of habitat and food supply (Ernvest, 2004).

Fish are one of the most widely distributed organisms in an aquatic environment and being susceptible to environmental contamination may reflect the extent of the biological effects of environmental pollution of water (Pant, et al., 1987).

2,4, Dichlorophenoxyacetic acid, commonly known as 2,4,D is a post-emergent, selective control herbicide used to control broad leaf weeds in agriculture, forestry and in home lawns and gardens. Worldwide, it is the most extensively used herbicide. 2,4,D belongs to the phenoxy class of herbicides, and is produced in a variety of forms, including: acid, salt, amine and ester (EPA U.S. 2005).

This research Aim to study effect of herbicide 2-4 D amine on the morphological and behaviors character on the fish Cyprinus carpio Linnaeus (common carp).

Materials and Methods

200 fish were collected from fish of different ages of common carp from Lake Euphrates, divided into two categories these fish depending on the age as it was the first age group ranging between (45-90) days, while the second category was age them between (90-150) days. Each category is divided into three replicates as well as a sample control and placed in Plastic tanks measuring 1.5 X 0.5 X 0.5 and by ten members in each basin. Plastic basins filled with fresh tap water before putting the fish and left to thunderous 48 hours to ensure depletion of chlorine from it and then placed the fish.

I left the fish under laboratory conditions has been provided with air using the air pumps small, such as those used in the Fish Accessories to ensure adequate supply of dissolved oxygen for fish at a temperature ranging between (24-28) c° for not less than three days in order to adapt to the conditions of the laboratory before used in the experiments.

Fish fed on diet brought from the lake itself was replaced every water basin (24) hours after feeding in order to maintain a healthy environment of oxygen was sterilized by pelvic potassium Bermnenat. Was prepared concentrations of the three herbicide 2.4. D Amine (2, 4 ,6)% dissolving a given weight of the pesticide in a liter of water and then took the concentrations of the three from the solution the record and by three replicates for each concentration and for the two categories both bug unit, while was considered the docks non-exposed control sample, and then determine the concentration of the pesticide is lethal to half the number of fish LC50 followers of the straight line equation.
Measured some of the properties physical and chemical for water basin, which is the temperature of the water and measured by the mercury thermometer with a normal gradient (0-100) ° C, and use the device pH meter, To measure the pH of the water basin after calibration standard intravenously, while the Electrical conductivity was measured by a device E.C. meter of the type HANNA She expressed the unity of the results (µs/cm) and using the DO meter factory by the company HANNA Also measured the amount of oxygen dissolved in the water has crossed the Yes Unit Results (mg / L), were calculated based on the value of the salinity values of electrical conductivity through the special equation (Mackereth, et.al., 1978), and depended on (Lind, 1979) Basin water hardness was measured and expressed the unity of the results (mg / L).

**Statistical analysis**

All data were expressed as means ±SD. least significant difference (LSD) was used for mean separation. The significant level was set at the probability level of P<0.05.

**Results**

Table (1) shows the values of the physical and chemical properties measured in the water basins. The results of the present study appear the clear influence of the herbicide 2-4 D amine on the fish, where there were appearance some morphological and behavioral signs on the fish when add the herbicide concentrations in the three basins, The first morphological signs that appeared was getting bleeding in Gills, as well as when you monitor the number of Strikes gill cover was found to be affected by increasing the concentration of the herbicide and this in turn affects the breathing and thus affect the metabolic and respiratory activity, And get a clear bleeding in the skin especially in the fish fin base and in addition to warp the neck of the tail and an increase in the fall of the scales on the body of the fish.

The terms of behavioral at the initial exposure, fish were alert, stopped swimming and remained static in position in response to sudden change in the surrounding environment. After some time they tried to avoid the toxic water with fast swimming and jumping. Faster opercula activity was observed as surfacing and gulping of air. In basins with high concentration of 2-4 D amine swimming of the fish was very erratic with vigorous jerks of the body. Their fins became hard and stretched of body muscles.

They secreted copious amounts of mucus from whole body continuously and soon thick layer of mucus was found deposited in the buccal cavity and gills. Body pigmentation was increased. Ultimately fish lost their balance and consciousness, engaged in rolling movement and became exhausted and lethargic. Lastly they remained in vertical position for a few minutes with anterior side or terminal mouth up near the surface of the water trying to gulp air and tail in a downward direction. Soon they settled at the bottom of the basin, and after some time their bellies turned upward and the fish died, while the opercula remained wide open exposing the gills.

Amount of Half lethal concentration LC50 for first category in all concentrations was (2.7938 %), while the second category in the all concentrations the LC50 was (1.5591 %), And the results of the present study showed affected the first category of *Cyprinus carpio* common carp fish more than the second category and this is evident by the results of statistical analysis that showed a correlation inverse association between the toxicity of the herbicide and between the ages of fish used. As shown by the results of statistical analysis no significant differences at the
level of probability (P < 0.05) between the two categories of fish used in the experiment.

The results showed that the higher concentration of herbicide is higher the mortality rate and this is clear from the results of statistical analysis which showed a positive correlation between both categories and the concentration of the herbicide and the proportion of deaths (toxicity).

**Table (1) Physical and chemical characteristics of the measured basin experiment**

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>First basin</th>
<th>Second basin</th>
<th>Third basin</th>
<th>Control basin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature</td>
<td>24 ± 1</td>
<td>25 ± 1</td>
<td>25 ± 1</td>
<td>24 ±1</td>
</tr>
<tr>
<td>pH</td>
<td>7.3 ± 0.4</td>
<td>7.6 ± 0.3</td>
<td>7.4 ± 0.2</td>
<td>7.4 ± 0.3</td>
</tr>
<tr>
<td>E.C.</td>
<td>673 ± 23</td>
<td>680 ± 13</td>
<td>667 ± 13</td>
<td>671 ± 17</td>
</tr>
<tr>
<td>D.O.</td>
<td>8.7 ± 1.9</td>
<td>9 ± 1.2</td>
<td>9.2 ± 0.8</td>
<td>9.5 ± 0.8</td>
</tr>
<tr>
<td>Salinity</td>
<td>0.471 ± 0.021</td>
<td>0.467 ± 0.033</td>
<td>0.483 ± 0.031</td>
<td>0.463 ± 0.041</td>
</tr>
<tr>
<td>Hardness</td>
<td>308 ± 10</td>
<td>310 ± 15</td>
<td>307 ± 15</td>
<td>293 ± 10</td>
</tr>
</tbody>
</table>

**Discussion:**

The use of chemical agents such as pesticides and herbicides has increased environmental risk to fish. The use of some fish for the purpose of monitoring and a sensitive indicators for environmental pollution (Seguini de Bravo, et al., 2005). Herbicides are considered hazardous pollutants to water and cause toxicity to fish and other aquatic invertebrates. This toxicity may end up with a man through the food chain. That the use of water in agriculture and industry requires the recycling and treatment of pollutants carried by before it reaches rivers and other sources, including drinking water and natural water (Abd-Algadir, et al., 2011).

In the present study found a clear effect of the herbicide 2-4-D on gills. This is due to the Gills usually appear to be one of the most damaged due to their direct contact with the toxic substances as they provide a very large interface between the external and internal environment of fish (Olivera Ribeiro, et al., 2002). This is not something strange, some studies have shown that fish exposed to pesticides cause damage to the gills tissue and changes in the rate of oxygen consumption. Shaffi, 1979 has been found, exposure of fish *Heteropneustes fossilis* to the pesticide *Hetachlor* causing damage to the gill tissue composition from its effect on epithelial cells and blood capillaries of the gill filaments and deposition of a thick layer of mucus led to block access oxygen to the blood. And thus failed to get the fish in need of oxygen. Marie, et al., 1998 As also noted in the occurrence of edema with separation of gill filaments and epithelial layer fabric in carp fish *Carpinus carpio* when exposed to lethal concentrations of the pesticide under the organic phosphoric Profenofos.

On the other hand Sastry, et al., 1988 found that exposure members of freshwater fish *Channa punctatus* to lethal concentrations and under the fatal cause of pesticide Sevin in a clear inhibition of enzyme activity Succinate dehydrogenase and Pyruvate dehydrogenase in the liver, muscles, and to an increase in enzyme activity Hexokinase and Lactate dehydrogenase, which indicates the rise in the rate of anaerobic metabolism in fish exposed to pesticide. Also reached Shaikila, et al., 1993 a similar conclusion when exposing the fish to the pesticide Sevin where noted increase in enzyme activity Acid phosphateasee who they saw as adaptive mutation in the tissue in order to meet its energy needs by promoting oxidative stress anaerobic, which may be due to lack of oxygen due to the low rate of entry into the blood by
gills a result of exposure to a pesticide, and from this we can deduce the intensity of the impact of pesticides on Gills.

There are many scientists who studied effect of various pesticides on fish and their Gomez, et al., (1998) Who has studied the impact of herbicide 2-4 D on a fish was found that the pesticide may have caused marked alteration of haematopoietic tissue, characterized by progressive swelling and cell necrosis, activation of the phagocyte system. There are a lot of research that showed that fish exposed to pesticides could lead to the killing through the inhibition of enzyme activity Acetyl Choline Esterase (AChE) when nerve receipts and muscle nerve receipts. Fulton and Key (2001) Has confirmed the existence of a link between the death of many species of fish and low enzyme (AChE) activity in the brain by more than 70% of these fish when exposure of pesticides. Herrera & Catap (2001) Noted that the exposure of young fish carp Cyprinus carpio concentration of 0.005 and 0.01 mg / l of the organic pesticide Azinophosethyl caused the decrease significantly in enzyme activity (AChE) in skeletal muscle, as shown by the study carried out by Philips, et al., (2002) about the effect of acute toxic pesticide Chlorpyrifos on fish on the type of Stizosadiom vitrum that the vulnerability of young fish to the pesticide caused the inhibition of enzyme activity (AChE) by up to 90% and that these young fish aged 60 and 90 days have lost their ability to survive when the ratio exceeded the inhibition of the activity of this enzyme to 71%.

During above exposure period the fish shows behavioral responses like increased opercular movement, mucous secretion, floating on the sides, fast swimming and etc. the abnormal behavior of the fish indicates the toxic effect of 2-4-D amine on Central Nerves System (CNS) and Cardiovascular System as suggested by Antychowicz, et al., (1979). and Hussiein, et al., (1996) suggested that these behavioral change were the result of decrease acetyl cholinesterase activity.

Also found in the current study that the fish the first category, the smallest most effective pesticide-lived fish, the second category of older and this study was matched with the study Jiraungkoorskul, et al., (2002) when studied Histopathological effects of Roundup, a Glyphosate Herbicide on the Nile tilapia Oreochromis niloticus.

Was a short trial period and this is due to the short life of 2-4 D herbicide in water, which quickly degrades or dissolves in water, affected by value of the acidic water. For this reason, we measured the pH of basins water used in the experiment to maintain a degree of parity so as not to degrade the pesticide. It was found that 2,4-D will change form and function with changes in water pH (Que Hee & Sutherland 1981). In alkaline (high pH; pH > 7) waters, 2,4-D will remain in a neutral molecular form, increasing its potential for adsorption to organic particles in water, and increasing its persistence (Wang, et al., 1994). 2,4-D is most likely to adsorb to suspended particles in muddy waters with a fine silt load (Que Hee & Sutherland, 1981), but little adsorption has been observed in the field (Halter, 1980).
References
- Halter, M. (1980). 2,4-D in the aquatic environment. Section II in Literature Reviews of Four Selected Herbicides: 2,4-D, dichlobenil, diquat & endothall. Shearer R., and M. Halter, eds.


