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## Effect of *Moringa oleifera* Crude Leaf Extract on Chlorpyrifos-induced Hepato-Toxicity in Wistar Albino Rats

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**Abstract** The effect of *Moringa oleifera* crude leaf extract on Chlorpyrifos-induced hepato-toxicity in Wistar albino rats was investigated. Sixteen (16) adult Wistar albino rats of weighing about 100g and 250g were divided into a group of four (4) of four (4) rats each. Group I served as the control and were given a standard feed while groups II, III and IV were given 200mg/kg, 100mg/kg and 100mg/kg of the feed respectively following different treatments with *Moringa* and Chlorpyrifos. All feed were administered in pelleted form for a period of 28days. On the 29th day, the animals were humanely sacrificed and blood collected for some biochemical analysis (liver enzymes marker). Body weight/feed consumption increased significantly ( $p < 0.001$ ) in group III when compared with groups I and IV. There was no significant difference ( $p > 0.05$ ) increase in group IV when compared with group II. The values of Aspartate transaminase (AST) and Alanine transaminase (ALT) were not significant ( $p > 0.05$ ) and decreased in group IV treated with *Moringa*/Chlorpyrifos compared with group II contaminated with Chlorpyrifos. The Alanine transaminase (ALP) level was significantly different ( $p < 0.001$ ) and increased in group II when compared with other groups. This study therefore showed that *Moringa oleifera* has the potential to increase body weight and offer some level of protection against Chlorpyrifos-induced hepato-toxicity.

**Keywords** *Moringa oleifera*, Chlorpyrifos, Body weight, AST, ALT, ALP

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### Introduction

The World Health Organization estimated that 80% of the populations of some Asian and African countries depend on traditional medicine for primary health care. *Moringa oleifera* Lam. (Moringaceae) is native to the southern foothills of the Himalayas in northwestern India and the most widely cultivated species of the genus *Moringa* [1]. It has become naturalized in many tropical countries of Africa, Arabia, South East Asia, Pacific Caribbean Islands and South America [2]. The common English names are: *Moringa*, drumstick tree, horseradish tree and benzoil tree. Locally, in Nigeria, it is known as ‘Zogale-gandi’ in Hausa, ‘Ewe igbale’ in Yoruba and ‘Okweoyibo’ in Igbo. It is also known as “Miracle tree”, [3]. *Moringa* is a versatile plant with high nutritive, agricultural, medicinal, domestic, industrial and environmental benefits. It is effective in combating malnutrition, especially among infants and nursing mothers [4]. The leaves, seeds and flowers have been reported to have good nutritive and medicinal value [5]. The seeds are consumed raw or roasted while the flowers are cooked in soups and resemble mushrooms [1]; the leaves are cooked as vegetables. The flowers and leaves are rich in vitamins A, B<sub>1</sub>, B<sub>2</sub>, B<sub>6</sub>, E and C. They are among the best sources of minerals. The plant has been implicated in the treatment or suppression of many degenerative diseases among many rural consumers [3]. In Northern Nigeria, *Moringa oleifera* is highly sourced as vegetable



because of its nutritional values, health promoting and disease-preventing properties possibly due to the presence of many phytochemicals [3].

Phytochemicals are produced in plants as protectants but researchers have demonstrated their role in the management of human diseases as well. The detailed account of the phytoconstituents may be useful for the synthesis of complex molecules of medicinal interest [6]. Various phytochemicals have been identified and isolated from different parts of *Moringa oleifera* [7].

The Chlorpyrifos pesticide is most commonly used by those working in the pest control to combat cockroaches and other household insects. A mixed cyclic compound, it is a complex molecules whose residues have longer lifetime than many Aliphatic or Vinyl derivatives and have inhibiting effect important in the nervous system enzymes and Cholinesterase enzymes (CHE), Cholinesterases. Inhibition of these enzymes causes accumulation of acetylcholine (Ach), which Interferes with the nervous muscle contact causing rapid twitch of voluntary muscles and leading to paralysis [8]. Chlopyriphos enters the body through the mouth, lungs and skin quickly passes after absorption from the intestine to the blood stream where it is distributed to the rest of the body parts [9].

The liver is one of the most rudimentary organs that engage in the biotransformation of nutrients; provide protection to the body against foreign agents, detoxification as well as the excretion of drugs and xenobiotics from the body [10]. Thus, it is requisite to protract liver strength for overall body's health and safety. Unluckily, environmental toxins, meager eating habits, alcohol and over-the-counter drug use are recurrent ill-treatments which can weaken the liver [11]. National Center for Health Statistics (NCHS) at the Centers for Disease Control and Prevention (CDC) considered chronic liver disease and cirrhosis as the 12<sup>th</sup> foremost basis of death which are asserting 30,000 lives in the United States per year [12]. Liver damage can be caused by many factors such as biological, autoimmune diseases, some drugs e.g.high dosage of paracetamol, antitubercular drugs, lethal compounds (such as carbon tetrachloride, thioacetamide, diethylenitrosamine, 4-Dglucosamine/lipopolysaccharides) and overdose of alcohol [13]. Therefore, the current study is aimed at investigating the effect of *Moringa oleifera* crude leaf extract on the lethal compound chlorpyrifos-induced hepatotoxicity on Wistar albino rats.

## Materials and Method

### Collection of Plant Material

*Moringa oleifera* leaves were obtained from Maiduguri metropolis. The plant was identified and authenticated by a plant taxonomist in the Department of Biological Science, University of Maiduguri, Nigeria.

### Experimental Animals

A total of sixteen adult Wistar strain albino rats of both sexes weighing between 100g and 250g were obtained from the Department of Biochemistry, University of Maiduguri, Nigeria. The animals were acclimatized under standard conditions having free access to feed and water *ad libitum*.

### Chemicals and reagents

All chemicals and reagents used in this study were of analytical standard. They included chlorpyrifos, liver enzymes parameter kits.

## Method

### Preparation of crude sample extract

Approximately 1kg of fresh tender leaves of *Moringa oleifera* leaves were washed, shed-dried at room temperature. The dried leaves were ground into a fine powder using mortar and pestle and stored in an air-tight container. About 250g of the dry powder was obtained.

### Experimental design

Sixteen Wistar albino rats were randomly divided into four (4) groups of four (4) rats each. They were treated as described below:

Group I (Normal Control): received standard diet (growers mesh).

Group II (200mg/kg): received 10% of Chlopyriphos (calculated based on the concentration) mixed with the standard diet.



Group III (100mg/kg feed): received crude *Moringa oleifera* leaf powder mixed with the standard diet.

Groups IV (100mg/kg feed): received crude *Moringa oleifera* leaf powder mixed with Chlorpyrifos and standard diet.

All the feed were administered in pelleted form after being weighed (20g per pellet) and the animals were fed as above for a period of four weeks (28days). The weight of the animals, feed consumed (subtracting feed remnants weight from initial feed weight) and drug consumption (Chlorpyrifos) were also obtained. On the 29th day, the animals were sacrificed and blood samples collected for some liver enzymes marker assay; Aspartate transaminase (AST), Alanine transaminase (ALT), Alkaline phosphatase (ALP).

#### Biochemical analysis

Serum alanine transaminase and aspartate transaminase were determined by the method described by [14] while serum alkaline phosphatase was carried out according to standard method [15].

#### Statistical analysis

The results were expressed as Mean  $\pm$  SEM (standard error of the mean) and Mean  $\pm$  SD (standard deviation). The statistical significance of the treatment effect was analyzed using the student's t-test statistics (Turkey HSD t-test). p values < 0.001 and P value > 0.05 were considered to be statistically significant and non-significant respectively.

#### Results

The mean weekly body weight, feed and drug consumption are shown in table 1. The result indicated a significant increase ( $p < 0.001$ ) in body weight and feed consumption in group III when compared with groups I, II and IV. The body weight also decreased in group IV when compared with group I though not significant  $p > 0.05$ . The drug consumption (Chlorpyrifos) in group IV showed no significance difference ( $p > 0.05$ ) when compared with group II.

Table 2 represents the mean value of the selected liver marker enzymes. The result showed that there was no significance difference ( $p > 0.05$ ) when group IV was compared with group II for the AST. No significant difference ( $p > 0.05$ ) was also recorded when group II was compared with group I. For ALT, the comparison between group I and groups II, III and IV showed no significance difference ( $p > 0.05$ ). When group II was compared with group I for the ALP, the increase was remarkably significant ( $p < 0.01$ ) however there was no significance difference ( $p > 0.05$ ) when group IV was compared with group II.

**Table 1:** Profile of the mean value of weekly body weight, feed and drug consumption of rats for 28days

Groups	Body weight (g)	Feed consumption (g)	Drug consumption (mg/ml)
I. Normal Control (Feed and water <i>ad libitum</i> )	140.78 $\pm$ 0.79 <sup>a</sup>	7.48 $\pm$ 2.22 <sup>a</sup>	Nil
II. Chlorpyrifos (200mg/kg feed)	139.88 $\pm$ 4.04 <sup>a</sup>	3.12 $\pm$ 1.09 <sup>a</sup>	0.62 $\pm$ 0.22 <sup>a</sup>
III. <i>Moringa oleifera</i> (100mg/kg feed)	188.47 $\pm$ 1.67 <sup>b</sup>	8.99 $\pm$ 3.03 <sup>a</sup>	Nil
IV. <i>Moringa oleifera</i> and Chlorpyrifos (100mg/kg feed)	133.91 $\pm$ 3.26 <sup>a</sup>	3.44 $\pm$ 1.48 <sup>a</sup>	0.69 $\pm$ 0.30 <sup>a</sup>

<sup>a</sup> = not significantly different ( $p > 0.05$ ), <sup>b</sup> = significantly different ( $p < 0.001$ )

All values are presented as Mean  $\pm$  Standard error of mean (SEM)

**Table 2:** Profile of the mean value of the liver marker enzymes for 28days

Groups	AST (IU/L)	ALT (IU/L)	ALP (IU/L)
I. Normal Control (Feed and water <i>ad libitum</i> )	146.60 $\pm$ 21.94 <sup>a</sup>	33.80 $\pm$ 4.49 <sup>a</sup>	69.00 $\pm$ 24.71 <sup>a</sup>
II. Chlorpyrifos (200mg/kg)	173.75 $\pm$ 21.84 <sup>a</sup>	40.00 $\pm$ 5.83 <sup>a</sup>	155.00 $\pm$ 16.2 <sup>b</sup>
III. <i>Moringa oleifera</i> (100mg/kg feed)	140.75 $\pm$ 11.12 <sup>a</sup>	33.25 $\pm$ 7.41 <sup>a</sup>	52.00 $\pm$ 17.06 <sup>a</sup>
IV. <i>Moringa oleifera</i> and Chlorpyrifos (100mg/kg feed)	155.00 $\pm$ 46.99 <sup>a</sup>	36.75 $\pm$ 15.56 <sup>a</sup>	99.67 $\pm$ 18.01 <sup>a</sup>

<sup>a</sup> = not significantly different ( $p > 0.05$ ), <sup>b</sup> = significantly different ( $p < 0.001$ )

All values are presented as Mean  $\pm$  Standard error of mean (SEM)



## Discussion

The result of the body weights and feed consumption of the rats for the period of four weeks revealed a significant increase ( $p < 0.001$ ) in group III compared to group I and group IV and also group III compared to group I. The increase in the body weight of rats might be unconnected with the fact that *Moringa oleifera* is rich in amino acids, vitamins and minerals particularly iron [16]. The significant increase in body weights of rats might also be attributed to captivity, where energy expenditure is minimal. This is in accordance with the findings of [17] and who also recorded increase in body weight of rats.

The liver is a vital organ in vertebrates and other animals. It is used in the elimination and detoxification of harmful biochemical waste products and toxins. It plays key role in the synthesis of biochemicals that are very vital in body metabolism. The hepatic function of the rats was assessed by serum ALT, AST and ALP. ALT and AST are cytosolic enzymes used to assess hepatocellular injury due to their increase release in to the circulation following damage to the hepatocytes. The significant drop in the levels of these enzymes due to *Moringa oleifera* treatment group (group IV) when compared to (group II) can be attributed to the anti-inflammatory properties of the plant. The anti-inflammatory activity can be traced to flavonoids like quecetin and phenolic acid present abundantly in the leaves of *Moringa oleifera* [18]. These compounds have documented anti-inflammatory properties [19].

The non significant ( $p > 0.05$ ) effect of *Moringa oleifera* and alkaline phosphatase (ALP) is an indication that the treatments have no untoward effect on the rats. This is in agreement with the findings of [20] who observed no effect of *Moringa oleifera* on alkaline phosphatase effect on the health status of rabbits. Alkaline phosphatase is present in all the tissues throughout the body, but is particularly concentrated in the liver, bile duct, kidney, bone and the placenta. It is therefore not a specific liver marker.

## Conclusion

Conclusively from our findings, it has been revealed that exposure to chlorpyrifos has some level of hepato-toxic effects as shown in the decrease in weight, elevated level of liver enzymes and also the ameliorately effect following *Moringa oleifera* supplementation. *Moringa oleifera* being rich in bioactive compounds may therefore be responsible for the pharmacological activity associated with it.

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