



RESEARCH ARTICLE

STUDY ON EFFECT OF CHLORPYRIFOS ON SPERM COUNT AND HEAD MORPHOLOGY IN MALE *Mus musculus*

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ARTICLE INFO

Article History:

Received 20th February, 2024

Received in revised form

25th March, 2024

Accepted 14th April, 2024

Published online 30th May, 2024

Key words:

Chlorpyrifos, Gonadotoxicity,
Mus musculus, Sperm count,
Abnormalities.

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ABSTRACT

Herbicides, Insecticides or pesticides etc may play a role in the development of adverse effects in human and animal bodies. Data concerning its mutagenic and carcinogenic properties have conflicting. In the present study, Effect of Chlorpyrifos on Sperm count and Sperm head abnormalities. Adult Swiss albino mice of the same age group were selected for the research experiment and were divided into three groups (n = 5) consisting of Group C, CPF-1 & CPF-2. Group C (Control Group) dosage of 1 ml/per day Distilled water (DW) orally for 6 weeks. Group CPF-1 (Lower dose Group) dosage chlorpyrifos in a dose of 10mg/kg body weight/per day orally for 6 weeks. Group CPF-2 (Higher dose Group) dosage chlorpyrifos in a dose of 20 mg/kg body weight/per day orally for 6 weeks. After the completion of dose duration, the experimental animals were euthanized by cervical dislocation and exposed both caput and cauda epididymis for the analysis of sperm count and sperm head abnormalities. The data obtained from the control and treated group are expressed as Mean% $\pm$ SE and ANOVA was used to determine by SPSS Software and the level of significance is $p < 0.05$. The frequency of sperm head abnormalities was significantly higher than that found in the control group. The frequency of sperm head abnormalities was found 3.09 ± 0.99 (Control), $6.92 \pm 1.46^*$ (CPF-1), and $14 \pm 2.00^{**}$ (CPF-2) and it is a significant difference between to control and treated groups at $p < 0.05$. The mean value of the sperm count was 157.02 ± 2.68 (Control), $82.1 \pm 2.81^*$ (CPF-1) and $46.5 \pm 2.78^{**}$ (CPF-2). The result show that in both CPF-1 & CPF-2 were significantly decrease the sperm count Hence, results indicate the potential of chlorpyrifos to be gonadotoxic.

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Citation: Manisha Rani and S.K. Singh 2024. "Study on effect of Chlorpyrifos on Sperm count and Head morphology in Male *Mus musculus*". International Journal of Current Research, 16, (05), 28340-28342.

INTRODUCTION

The environment is increasingly being influenced by the presence of waste and byproducts of natural and anthropogenic origin. Anthropogenic pollution is evidenced in a large number of chemicals, such as Metals, Heavy Metals, Drugs, Food additives, Herbicides, Insecticides or pesticides etc which would be largely responsible for damage and alterations at morphological and genetic levels in several species (ATSDR, 2000). The most popular pesticides used globally to fumigate crops and plants are organophosphates. Organophosphorus pesticides (OPs) are one of the most potent pesticides. Among the OPs Chlorpyrifos (CP) [O, O-diethyl-O-(3,5,6-trichloro-2-pyridinyl)-phosphothioate] is one of the oldest, wide spectrum insecticide, used in public health and residential to kill a variety of insects, as well as for the control of the vectors of diseases. Several studies have reported that CPF has the potential to induce Reactive Oxygen Species, lipid peroxidation, and enzyme inactivation of cellular membranes through various biochemical processes

(Gupta *et al.*, 2010, Uchendu, 2012) and the metabolites produces by the oxidation of CP show a pattern of induction of chromosome damage and has been related to gonadotoxicity and genotoxicity (Li *et al.*, 2015 & Sud, 2020). Therefore, the aims and objectives of the research work was to estimate and determine the effects of Chlorpyrifos (CPF) on sperm count and head morphology in caput and cauda epididymis of Male *Mus musculus*.

MATERIALS AND METHODS

Experimental animals: For the research experiment, 15 adult Male Mice (*M. musculus*) of same age and average weight of 25- 30-gram Body Wt. were taken from the animal house of the University Department of Zoology, T. M. B. U., Bhagalpur (Bihar). All the experimental animals were kept in a polypropylene cage under hygienic conditions in a well-ventilated room. For the research experiment and administration of dose experimental animals were divided into equal number of mice in three groups one group was

considered the control (C) two were considered treated (CPF-1 & CPF-2) groups.

METHODOLOGY

Controls groups of experimental animals were fed 1ml of D.W and treated groups were fed 1 ml of CPF orally with the help of gastric gavage. After the completion of treatment duration (6 weeks) both control and treated groups *M. musculus* was euthanized by cervical dislocation and cauda and caput epididymis were exposed into watch glass. Each caput and cauda epididymal part minced with forceps and needle in 1 ml of normal saline and were sieved by a metallic filter to avoid the tissue debris in seminal content. Permanent slides of sperm head morphology were prepared by the methodology of Wyrobek *et al.*, (1983). Approx. 5000 sperm cells were screened for each group. The study of sperm head morphology under microscope and sperm count was done by Neubauer haemocytometer method (Eliasson, 1975).

STATISTICAL ANALYSIS

Statistical analysis: Data were analyzed using S.P.S.S. software. In each experimental variant obtained data from the control and treated groups are expressed as Mean $\pm$ SEM and for the comparison of data between the control and treated groups ANOVA was used to determine at significant level $p < 0.05$ & 0.01 .

RESULT AND DISCUSSION

In the present research oral feeding of chlorpyrifos (CPF) cause significant decline in sperm counting and increase of sperm head abnormalities. The number of sperms was 157.02 ± 2.68 (control), $82.1 \pm 2.81^*$ (lower dose), and $46.5 \pm 2.78^{**}$ (higher dose) million cells/ml in groups C, CPF-1 and CPF-2 respectively (Table-1, Figure -1) and Different types of abnormalities were observed including pinheaded, banana shaped, double headed, hammer shaped, coil shaped, tail less, funnel shaped, ring shaped in treated group. The result of the sperm head morphological abnormalities was 3.09 ± 0.99 (control), $6.92 \pm 1.46^*$ (lower dose), and $14 \pm 2.00^{**}$ and CPF-1 and CPF-2 were significantly higher than control group (Table -2, Figure-2).

Table 1. Result of sperm count of *Mus musculus* treated with lower dose and higher dose CPF in 6 weeks of treatment duration

Experimental variant	Sperm counts ($\times 10^4$ Sperm / ml)
Control	157.02 ± 2.68
CPF-1	$82.1 \pm 2.81^*$
CPF-2	$46.5 \pm 2.78^{**}$

*, **, indicates the significant, and highly significant difference to control, CPF-1 and CPF-2 respectively. Notes – values expressed as Mean $\pm$ SEM and p value were analysed using ANOVA Test for multiple comparison.

Table 2. Result of Effect on sperm head morphology of *Mus musculus* treated with lower dose and higher dose CPF in 6 weeks of treatment duration

Experimental variant	Mean % $\pm$ SE
Control	3.09 ± 0.99
CPF-1	$6.92 \pm 1.46^*$
CPF-2	$14 \pm 2.00^{**}$

*, **, indicates the significant, and highly significant difference to control, CPF-1 and CPF-2 respectively. Notes – values expressed as Mean % $\pm$ SEM and p value were analysed using ANOVA Test for multiple comparison.

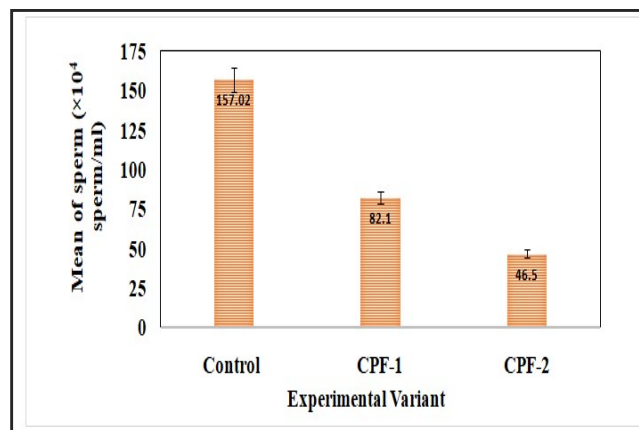


Figure 1. Histogram showing Mean of sperm count in caput epididymis of *Mus musculus* in 6 weeks of treatment duration

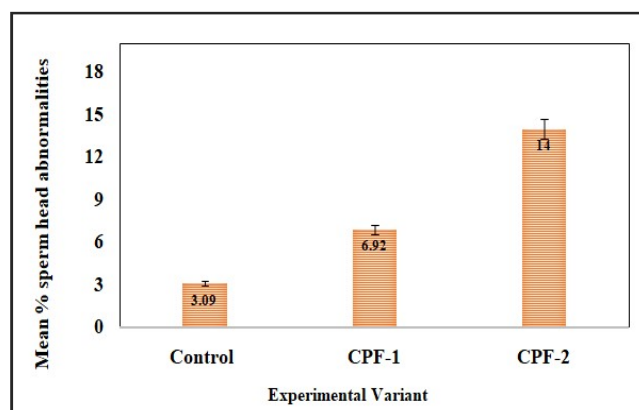


Figure 2. Histogram showing Mean % of sperm head abnormalities of *Mus musculus* in 6 weeks of treatment duration

CONCLUSION

From the obtained experimental result data, it is concluded that the pesticides chlorpyrifos produces a gonadotoxic effect on germinal cells it reduces the number of sperm cells and increasing the sperm head abnormalities in male Swiss albino mice (*Mus musculus*). The result revealed that health and environmental risks by pesticides discharge into the waterbodies and soil.

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