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TESTICULAR OXIDATIVE STRESS AND BIOCHEMICAL PERTURBATIONS INDUCED BY LAMBDA CYHALOTHRIN AND THE PROTECTIVE EFFECTS OF TAURINE IN RATS

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ABSTRACT

Lambda-cyhalothrin is a type II pyrethroid insecticide and may cause serious environmental pollution and health problems. Taurine, 2-amino ethanesulphonic acid is an essential amino acid, plays important roles in numerous physiological functions. The present study was conducted to evaluate the adverse effects of lambda cyhalothrin on the reproductive system of male Wistar rats and also to evaluate the protective role of taurine under these conditions**.** In the study sexually mature male rats were orally received lambda cyhalothrin at two different dose levels (s10.83mg/kg body wt. i.e. 1/7th of LD_{50} value and 15.17mg/ kg body wt. i.e. 1/5 of LD_{50} value) for 14 consecutive days along with pre-treatment of taurine(50mg/kg body wt.). A significant decrease in sperm motility, seminal fructose concentration, reduced glutathione and increase in testicular acid phosphatase, glutamate -oxaloacetate transaminase and glutamate-pyruvate transaminase activity, malondialdehyde and oxidized glutathione were observed in lambda cyhalothrin intoxicated rats. However, pretreatment with taurine significantly restored the above said parameters close to the normal level. The results disclose the toxic effect of lambda cyhalothrin on male reproductive system of rat and also point towards the beneficial influences of taurine in reducing the harmful effects of lambda cyhalothrin in this situation.

KEYWORDS: Lambda-cyhalothrin; Taurine; Testicular; Oxidative stress; Biochemical parameters.

INTRODUCTION

Although in terms of prosperity, green revolution has brought a windfall to the farmers in the field of agriculture, it is now showing its side-effects in the form of large-scale environmental degradation with chemical pesticides leading to the contamination of water, food and air. There has been indiscriminate use of pesticides in agriculture as well as in household and this is one of the major health issues in both developing and developed countries.[1] It disrupts the male reproduction in case of both humans and wildlife. Hence, testicular toxicity is of serious concern because a large number of pesticide is affecting badly the testicular functions in experimental animals.[2,3] The biggest group of broad-spectrum organic insecticides is synthetic pyrethroids which is used in agricultural, veterinary and household applications.[4] Lambda-cyhalothrin (LCT) is a type II pyrethroid, used mainly to control insect pest in agriculture, public health, homes and gardens. LCT is found in vegetables and fruits[5], milk and blood of dairy cows[6] as well as in cattle meat.[7] Placental transfer of LCT has been detected in goats.[8] It has also been noticed that LCT brings significant genotoxic and cytotoxic effects on human lymphocytes cultured *in vitro*[9], a dose dependent chromosomal aberrations in mice[10] and also changes in rabbit peripheral blood lymphocytes.[11] It has already been seen that LCT causes hepatotoxicity.

Taurine (TA), a sulphur comprising β-amino acid is present in most animal tissues. It is required for the normal functioning of different organs.[12] Biosynthesis and dietary intake from meat and mostly sea food[13] are the sources of TA in the body. Furthermore, TA prevents toxin-mediated hepatic injuries by minimizing oxidative stress, strengthening mitochondrial function and regulating cytoplasmic and mitochondrial calcium homeostasis.[14] Apart from it, TA also brings nephroprotective effects, perhaps because of its antioxidant and membrane stabilization effects.[15,16]

It is our hypothesis in the current study that LCT administration in the male Wistar rats may bring hazardous effects on sperm motility which includes testicular biochemical and oxidative stress parameters. Moreover, we also hypothesized that TA may decrease the harmful effects of LCT on testicular biomarkers in the rats based on its bioprotective effects on testis. This study has quite significance as pyrethroid is priority chemical for risk assessment because of their multiple organ toxicity and there is a strong possibility of the co-exposure of humans and other living organisms in the ecosystem to LCT. So, the current study has been done to assess the testicular toxicity in terms of biochemical alterations in

testis and oxidative stress caused by LCT in male rat and the pretreatment of taurine on biochemical parameters in LCT intoxicated male Wistar rats.

MATERIALS AND METHODS

Chemicals and reagents

Lambda cyhalothrin 5% emulsifiable concentrate (EC) was procured from RPC Agro Industries, Kolkata. Taurine was purchased Sigma Aldrich Inc. USA. All other chemicals used were of analytical grade and were obtained from Merck India Ltd, Himedia India Ltd, etc.

Animal mode

Healthy Wistar albino male rats (*Rattus norvegicus*) (weighing 130-150 g) were chosen for this experiment on the basis of easy availability and handling under normal laboratory conditions. The animals were fed on a standard diet and water *ad libitum* and kept in a temperature controlled environment (20–22◦C) with an alternating cycle of 12 h light and dark. The experiments reported here were approved by the Institutional Animal Ethical Committee, registered under CPCSEA. All animal treatment and surgical procedures were carried out according to the relevant laws and guidelines of the CPCSEA.

Treatment protocol

Lambda cyhalothrin 5% emulsifiable concentrate (EC), a commercial formulation was used for this experiment. It was in emulsion form and dilutions were prepared in distilled water to obtain the test concentrations which were calculated from the percentage of the active ingredient of commercial formulation of lambda cyhalothrin following the oral LD_{50} 75.85-mg/kg body weight in male rats.[17] During the experimental duration, body weights were daily recorded and the doses of the tested compounds were prepared accordingly.

After acclimatization period for one week under laboratory conditions, rats were randomly divided into six groups each containing six rats. Group I: Distilled water control (no treatment). Group II: Taurine control (taurine at a dose level of 50mg/kg body wt.).[18,19] Group III: Lambda cyhalothrin low dose (lambda cyhalothrin at a dose level of 10.83mg/kg body wt.).[17] Group IV: Taurine (50mg/kg body wt.) + lambda cyhalothrin low dose (10.83mg/kg body wt.). Group V: Lambda cyhalothrin high dose (lambda cyhalothrin at a dose level of 15.17mg/kg body wt.) and Group VI: Taurine (50mg/kg body wt.) + lambda cyhalothrin high dose (15.17mg/kg body wt.). The route of application chosen for the study

was daily oral gavage for 14 consecutive days. At the end of the treatment schedule, the animals were fasted overnight, anesthetized using pentobarbital sodium (35mg/kg) and sacrificed by cervical dislocation on 15th day.

Sample collection

After sacrifice, testis from control and treated rats were collected and immediately stored at -80ºC until analysis. Epididymis were collected and washed immediately for sperm collection.

Determination of Testicular index

Testes of sacrificed male Wister albino rat were dissected from its body and all fats were removed from it. Then their weights were taken to calculate testicular index using the following formula:

$$\text{Testicular index} = \frac{\text{Testicular weight}}{\text{Body weight}} \times 100$$

Sperm motility

Sperm motility was evaluated by the counting method of the motile and non -motile spermatozoa under microscope and expressed as the percent motility.[20]

Measurement of seminal fructose concentration

In a centrifuge tube, 1ml of diluted seminal plasma (five time dilution was done by mixing 0.1ml of seminal plasma with 4.9ml of distilled water) was mixed with 0.3ml of 1.8gm% $ZnSO_4$ and 2ml of 0.1M NaOH. To get the supernatant, the above mixture was centrifuged at 2000g after 15min. Then seminal fructose concentration was estimated by taking 0.5ml of supernatant as sample, 0.5ml of 0.14mM and 0.28mM fructose solutions as two standards and 0.5 ml of distilled water as blank and added 0.5 ml of indole reagent, 5 ml of concentrated HCl to each test tube. The test tubes were incubated at 50^0C for 20 min and were cooled in ice water and then in room temperature.[21] The reading was noted at 470 nm in spectrophotometer (UV-245 Shimadzu, Japan).

Assay of acid phosphatase

The acid phosphatase activity was measured in an acetate buffer at pH 4.5 using *p*-nitrophenol phosphate as a substrate.[22] Amount of PNP liberated was measured spectrophotometrically at 420 nm.

Activity of testicular glutamate-oxaloacetate transaminase (GOT) and glutamate-pyruvate transaminase (GPT)

To prepare sample for GOT, 1 ml of buffer substrate (prepared by taking 2.66gm aspartic acid, 60mg α-ketoglutaric acid and 20.5ml 1N NaOH and volume was made upto 100ml by 0.1M phosphate buffer, pH7.4) and for the preparation of sample for GPT, 1 ml of buffer substrate (containing 1.78 gm DL-alanine, 30mg α-ketoglutaric acid, 20 ml 0.1M phosphate buffer and 1.25ml of 0.4N NaOH) was taken separately and allowed to wait for 5min at 37^0C. Then 0.2ml tissue homogenate (20mg testis tissue/ml phosphate buffer) was added to each sample and incubated for 60 min at 37^0C. To prepare standard, 0.2 ml of working standard (200µM/100 ml) was received in a test tube and 0.8ml of buffer substrate was added. For blank, 1.0 ml of buffer substrate was taken. In each of sample, standard and blank test tubes, 1ml of DNPH solution were mixed and allowed to wait for another 20 min. Then 10ml of 0.4N NaOH was mixed and allowed to stand for 10 minutes. Readings were measured in spectrophotometer (UV-245 Shimadzu, Japan) at 520 nm.[23]

Estimation of lipid peroxidation

Quantitative measurement of lipid peroxidation was performed in testicular tissue homogenate (20mg/ml in phosphate buffer) according to the method of Ohkawa et al.[24] based on the formation of thiobarbituric acid reactive substances (TBARS) in term of malondialdehyde (MDA) formation. The absorbance of supernatants was taken at 535nm in spectrophotometer (UV-245 Shimadzu, Japan).

Determination of reduced glutathione and oxidized glutathione

Testicular reduced glutathione (GSH) estimation was performed by the method of Griffith.[25] The GSH was expressed as µg/mg protein. The reaction mixture contained 200µl of tissue homogenate (20mg/ml in phosphate buffer), 100 µl of sulfosalicylic acidand centrifuged for 10 min at 3000 rpm. After addition of 1.8 ml of DTNB with the supernatant, reading was taken at 420nm.

The oxidized glutathione (GSSG) was measured according to the method of Griffith[26] using 2µl of 2-vinyl pyridine, 250µl of sulfosalicylic acid (4 gm %) and 2 ml of DTNB (4mg %). The reading was taken at 412 nm. The GSSG was expressed as µg/mg protein.

Assay of of glutathione-S-transferase

Glutathione-S-transferase (GST) activity was quantified spectrophotometrically.[27] Briefly in a cuvette, 0.1 ml of tissue homogenate(20mg/ml in phosphate buffer), 2.8ml of 20mg/ml in phosphate buffer, 0.1ml of GSH and 50µl of 60mM CDNB were taken, mixed and reading was noted at 340nm. The values were expressed in µmol CDNB conjugate formed/min/ per milligram protein.

Statistical analysis

All the parameters were assayed in triplicate manner. The data was expressed as Mean±SEM. The differences between the means of each group were tested using a one-way ANOVA test (using a statistical package, Origin 6.1, Northampton, MA). $P<0.05$ was considered to indicate a statistically significant difference.

RESULTS

Effect on testicular index

The testicular index was decreased significantly ($p<0.001$) in a dose dependent manner in LCT treated group which was elevated significantly ($p<0.01$) by the pre-treatment of taurine(fig.1).

Sperm motility

Significant decrease ($p<0.001$) in sperm motility was observed in the LCT treated group compared to the control group (fig.2). Pre-treatment with taurine significantly ($p<0.001$) increased the sperm motility to a good extent.

Effect on seminal fructose concentration

Seminal fructose concentration was found to be decrease significantly ($p<0.001$) ina dose dependent manner in LCT treated group, compared to control (fig.3). Pre-supplementation of taurine rescues it from LCT induced toxicity.

Assay of acid phosphatase activity

Acid phosphatase activity was significantly elevated($p<0.01$) in the testis after LCT treatment, in comparison to the controls (fig.4). Pretreatment of taurine reduced it but not significantly.

Estimation of testicular glutamate-oxaloacetate transaminase (GOT) and glutamate-pyruvate transaminase (GPT) activity

Testicular glutamate-oxaloacetate transaminase (GOT) and glutamate-pyruvate transaminase (GPT) activity in LCT treated rat testis were found to be increased compared to that of control. Taurine restored these parameters towards more or less control level (fig.5, 6).

Lipid peroxidation and glutathione content

A significant increase ($p<0.001$) in MDA concentrations was observed in rat testes exposed to LCT pyrethroid in a dose dependent manner (fig.7).

On the other hand, a significant decrease ($p<0.001$) in GSH content (fig.8) with asignificant dose dependent increase in GSSG level in LCT treated rat testes was seen (fig.9).

Protective effects of taurine were evident after the pretreatment of taurine followed by LCT, where the alterations in the lipid peroxidation and glutathione content were restored close to the normal level.

Glutathione-S-transferaseactivity

From the result it is well-established that LCT treatment showed significant inhibition of antioxidant enzyme glutathione-S-transferase (GST) activities. The activity of the GST was better in taurine pre-treated group (fig.10).

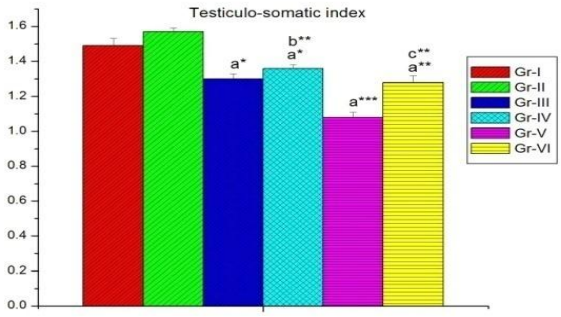

Figure1 shows the effect of taurine on testiculo-somatic index in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level of significance (* indicates $p<0.05$, ** indicates $p<0.01$, * indicates $p<0.001$).**

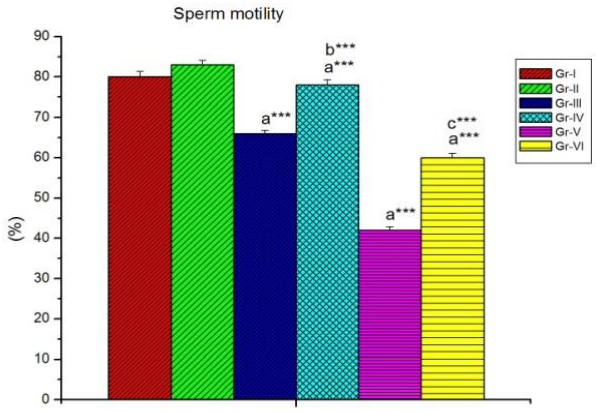

Figure2. The effect of taurine on sperm motility in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level of significance (* indicates p<0.001).**

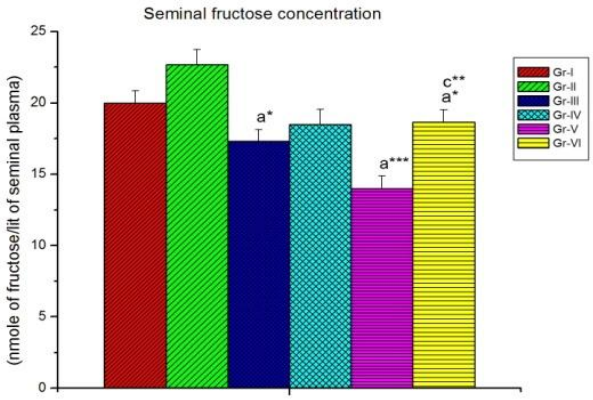

Figure 3 shows the effect of taurine on seminal fructose concentration in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level of significance (* indicates p<0.05, ** indicates p<0.01, * indicates p<0.001).**

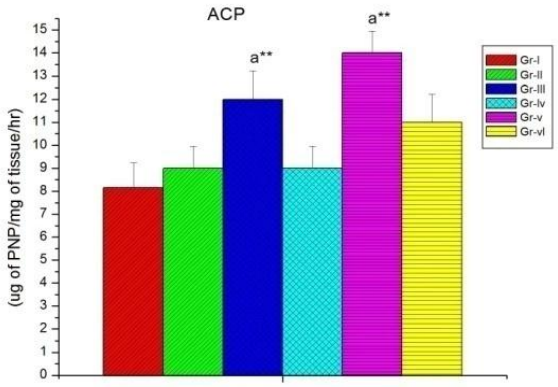

Figure 4. The effect of taurine on acid phosphatase activity in LCT induced male rat . Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level of significance (indicates p<0.01).**

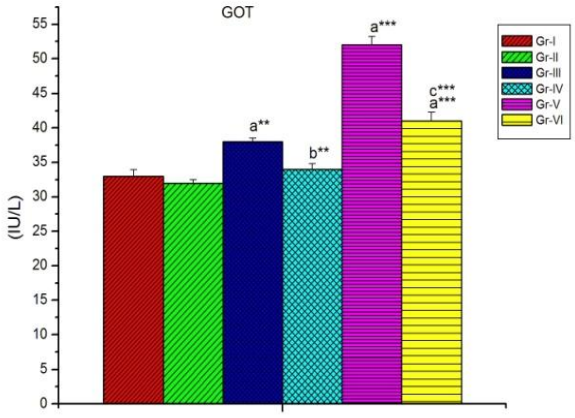

Figure 5 shows the effect of taurine on testicular glutamate-oxaloacetate transaminase (GOT) in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level ofsignificance (indicates p<0.01, *** indicates p<0.001).**

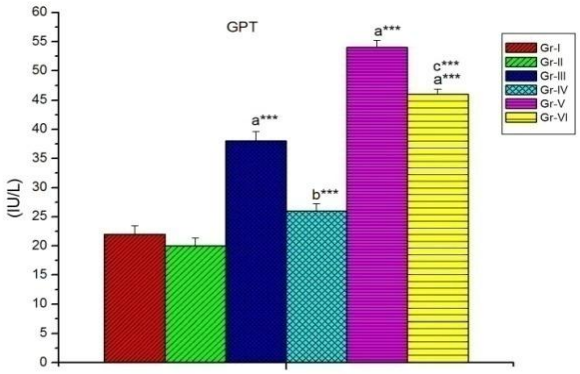

Figure 6. The effect of taurine on testicular glutamate-pyruvate transaminase (GPT) in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represents the different level ofsignificance (* indicates p<0.001).**

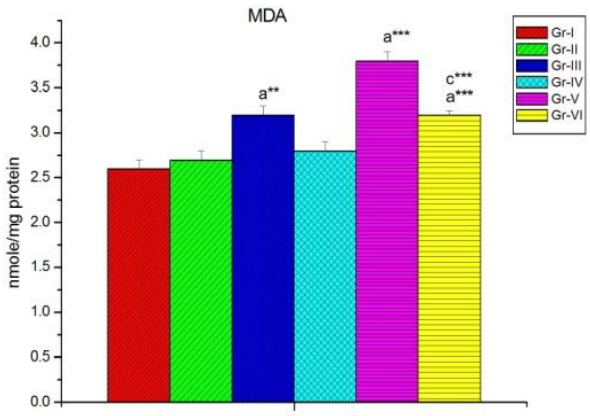

Figure 7 shows the effect of taurine on testicular malondialdehyde content in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level of significance (indicates p<0.01, *** indicates p<0.001).**

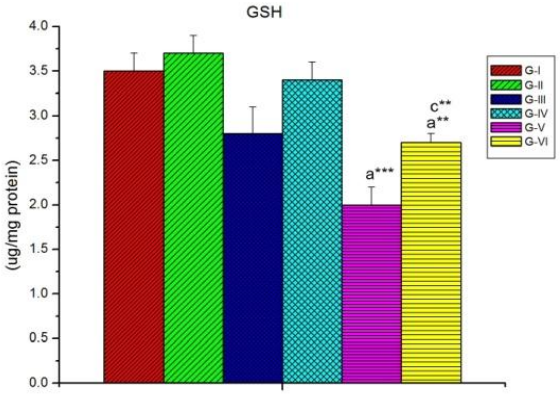

Figure 8. The effect of taurine on testicular reduced glutathione level in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represents the different level of significance (indicates p<0.01, *** indicates p<0.001).**

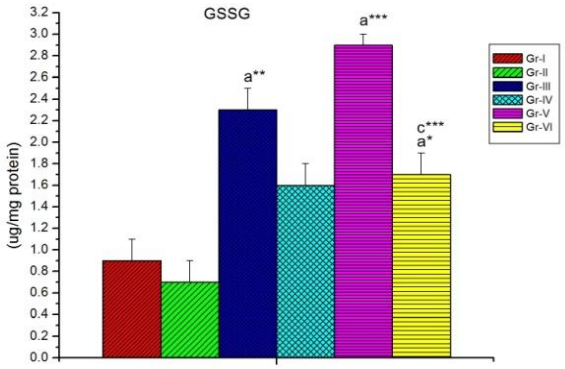

Figure 9 shows the effect of taurine on testicular oxidized glutathione in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level of significance (* indicates p<0.05, ** indicates p<0.01,* indicates p<0.001).**

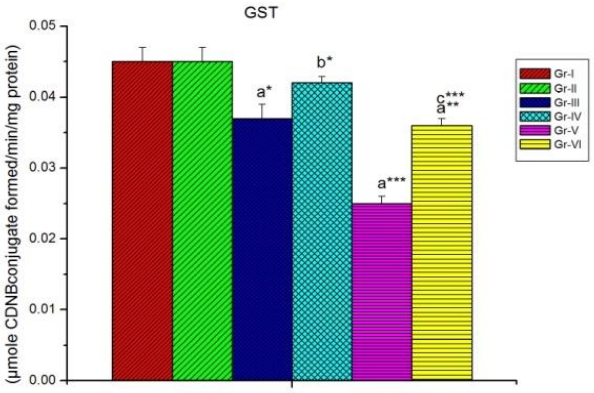

Figure 10 shows the effect of taurine on testicular glutathione –S-transferase activity in LCT induced male rat. Results are expressed as Mean±SEM. Analysis is done by one-way ANOVA followed by multiple comparison two-tail t-tests. Superscript a, Group-I versus all other groups; Superscript b, Group-III versus Group-IV; Superscript c, Group-V versus Group-VI. Asterisks represent the different level of significance (* indicates p<0.05, ** indicates p<0.01, * indicates p<0.001).**

DISCUSSION

The current study has been done to evaluate the harmful effects of lambda cyhalothrin (LCT) on the reproductive system of male Wistar rats, and to assess the controlling role of taurine under the toxic situation. The testicular index relies on both testicular weight and body weight. Reduction in the testicular weight on LCT exposure is perhaps due to decreased tubule size, reduced number of germ cells and enlarged spermatids.[28] The testicular index reduced by LCT intoxication successfully raised by the treatment of taurine. Perhaps, this is due to the preventive role of taurine on testicular damage. Decline in sperm motility after oral administration of LCT was either by androgen deprivation effect of the pyrethroid or by low spermatogenesis. Another possibility was enhanced reactive oxygen species ROS production by pyrethroid exposure. Similar results were also reported earlier.[29-31] Taurine pretreatment significantly improves the sperm parameters, indicated the important role of taurine to improve the semen quality either by its effect on stimulation of testosterone secretion[32] or by maintaining the testicular homeostasis.

The useful biochemical indicators estimated in this experiment reveals the significant alteration in the seminal fructose content, the ‘marker’ for the functioning of seminal vesicle[33], testicular acid phosphates, testicular glutamate pyruvate transaminase(GPT),

glutamate oxaloacetate transaminase(GPT) activity, testicular malondialdehyde level and glutathione-S-transferase (GST) in LCT intoxicated rat compared to control. Fructose is the primary secretory product of seminal vesicle that offers nutrients for the semen also important for sperm motility and stability of sperm chromatin. Reduced fructose content indicated that the secretary ability of the seminal vesicle was hampered by the pyrethroid treatment[34], which adversely affect the nutritive potentials for the semen in turn affect sperm motility. In this experiment seminal fructose concentration was found to be significantly lower in the LCT intoxicated groups compared to the rats in the control group, with the exception of taurine treated groups. This type of result clearly indicates LCT induced testicular damage. Testicular acid phosphatase activity was an important marker to assess the male reproductive toxicity. It takes a role in the cell metabolic process, autolysis and also in the differentiation including many related processes. Dilatation of blood capillaries in between seminiferous tubules is the result of acid phosphatase enzyme activity. The increase in acid phospahatase enzyme activity could be explained on the basis of enhancement of cell membrane permeability with an interruption in the transphosphorylation process as a result of cellular degeneration.[35] Increased testicular GOT and GPT levels indicates towards the LCT induced testicular injury.[36] Taurine pretreatment significantly improves the sperm parameters, fructose content and normalize the GOT, GPT levels. Testicular acid phosphatase activity was improved but not significantly.

During pyrethroid metabolism, excess reactive oxygen species (ROS) were produced which caused oxidative stress in pyrethroid induced animals.[37] Lipid peroxidation is believed to be one of the main markers of ROS-mediated damage. In our experiment malondialdehyde (MDA) level in LCT treated group was found to be significantly higher than that of control. Excessive production of reactive oxygen species (ROS) is considered to be also associated with the inhibition of endogenous antioxidant defense system that are also responsible to neutralize the toxic effects of these free radicals by giving electrons to these toxic species. Keeping in mind that GSTs are detoxifying enzymes which effectively catalyzes the conjugation of a variety of electrophilic substrates to the thiol group of GSH to create less toxic forms.[38] The significant reduction of GST activity shows insufficient detoxification process in LCT intoxicated male rats. Yamamoto and Yamashita concluded that detoxification system created by GST may be reduced by the presence or action of ROS.[39] Co-administration of taurine notably improved these abnormalities to a great extent. It decreased lipid peroxidation either by scavenging or reducing reactive oxygen species. In

various studies, the invigorating effects of taurine on endogenous antioxidants were already confirmed. On the whole, the results not only have given strong support for the toxic effect of LCT, but have also indicated the preventive role of taurine in this condition.

CONCLUSION

From the above discussion it can be inferred that lambda cyhalothrin creates testicular toxicity, lipid peroxidation, antioxidant insufficiency in male rats and co-supplementation of taurine has therapeutic effects on lambda cyhalothrin-induced male reproductive toxicity.

CONFLICT OF INTEREST

Authors declare that there are no conflicts of interest.

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REFERENCES

1. Peter JV, Cherian AM. Organic insecticides. Anaesth Intensive Care. 2000; 28: 11-21.
2. Joshi SC, Mathur R, Gulati N. Testicular toxicity of chlorpyrifos (an organophosphate pesticide) in albino rat. Toxicology & Industrial Health. 2007; 23: 439–444.
3. Shalaby MA, El Zorba HY, Ziada RM. Reproductive toxicity of methomyl insecticide in male rats and protective effect of folic acid. Food and Chemical Toxicology. 2010; 48: 3221–3226.
4. Righi AD and Palermo-Neto J. Behavioral effects of type II pyrethroid cyhalothrin in rats. Toxicology and Applied Pharmacology. 2003; 191: 167–176.
5. Turgut C, Ornek H, Cutright TJ. Determination of pesticide residues in Turkey's table grapes: the effect of integrated pest management organic farming and conventional farming. Environmental Monitoring and Assessment. 2011; 173: 315–323.
6. Bissacot DZ, Vassilieff I. Pyrethroid residues in milk and blood of dairy cows following single topical applications. Veterinary and human toxicology. 1997; 39: 6-8.
7. Muhammad F, Akhtar M, Rahman ZU, Farooq HU, Khaliq T, Anwar MI. Multi-residue determination of pesticides in the meat of cattle in Faisalabad- Pakistan. Egyptian Academic Journal of Biological Sciences. 2010; 2: 19-28.
8. Oliveira C, Vassilieff VS, Vassilieff I. Residues and placental transfer of lambda-cyhalothrin in goats. J. Vet. Res. 2000; 4: 146-152.

9. Naravaneni R and Jamil K. Evaluation of cytogenetic effects of lambda-cyhalothrin on human lymphocytes. Journal of Biochemical and Molecular Toxicology. 2005; 19: 304–310.
10. Chauhan LKS, Chandra S, Saxena PN and Gupta SK. In vivo cytogenetic effects of a commercially formulated mixture of cypermethrin and quinalphos in mice. Mutation Research Genetic Toxicology and Environmental Mutagenesis. 2005; 587: 120–125.
11. Georgieva SV. Investigation of the cytogenetic effect of the insecticide karate on rabbit peripheral blood lymphocytes. Trakia Journal of Sciences, 2006; 4: 34–38.
12. Brosnan JT, Brosnan ME. The sulphur-containing amino acids: an overview. Journal of Nutrition. 2006; 136: 1636-1640.
13. Ito T, Schaffer SW, Azuma J. The potential use fulness of taurine on diabetes mellitus and its complications. Amino Acids. 2012; 42: 1529-1539.
14. Asha KK, Devadasan K. Protective effect of taurine on the mitochondria of albino rats induced with fulminant hepatic failure. Biomedicine & Preventive Nutrition. 2013; 3: 279-283.
15. Chesney RW, Han X, Patters AB. Taurine and the renal system. Journal of Biomedical Science. 2010; 17: 1-10.
16. Han X, Chesney RW. The role of taurine in renal disorders. Amino. Acids. 2012; 43: 2249-2263.
17. Sharma CD, Saxena NP & Sharma R. Assessment of clastogenicity of lambda cyhalothrin, A synthetic pyrethroid in cultured lymphocytes of albino rats. World Applied Sciences Journal. 2010; 8: 1093-1099.
18. Ozden S, Catalgol B, Gezginci-Oktayoglu S, Arda-Pirincci P, Bolkent S, Alpertunga B. Methiocarb-induced oxidative damage following subacute exposure and the protective effects of vitamin E and taurine in rats. Food and chemical Toxicology. 2009; 47: 1676-1684.
19. Cetiner M, Sener G, Sehirli AO, Eksioglu-Demiralp E, Ercan F, Sirvanci S, Gedik N, Akpulat S, Tecimer T & Yegen BC. Taurine protects against methotrexate-induced toxicity and inhibits leukocyte death. Toxicology and Applied Pharmacology. 2005; 209: 39-50.
20. WHO. Laboratory manual for the examination of human semen and sperm–cervical mucus interaction. Fourth edition. New York. Cambridge University Press: 1999.
21. Karvonen M.J. and Malm M. Colorimetric Determination of Fructose with Indol, Scandinavian. Journal of Clinical and Laboratory Investigation. 1955; 7: 305-7.

22. Vanha-Perttula T and Nikkanen V. Acid phosphatases of the rat testis in experimental conditions. Acta Endocrinol. 1973; 72: 376–390.
23. Goel BK. Routine biochemical tests. In: Mukherjee KL, editor. Medical Laboratory Technology, Vol.III. New Delhi: Tata Mc Graw-Hill publishing company Ltd, 1988; 985-1079.
24. Ohkawa H, Onishi N and Yagi K. Assay for lipid per-oxidation in animal tissue by thiobarbituric acid reaction. Analytical Biochemistry. 1979; 95: 351-358.
25. Griffith OW. Glutathione turnover in human erythrocytes. Journal of Biochemistry. 1981; 256: 4900-4904.
26. Griffith OW. Determination of glutathione and glutathione disulphide using glutathione reductase and 2-vinylpyridine. Analytical Biochemistry. 1980; 106: 207-12.
27. Habig WH, Pabst MJ & Jakoby WB. Glutathione-S-Transferase: the first enzymatic step in mercapturic acid formation. Journal of Biochemistry. 1974; 249: 7130–7139.
28. Sanhez LC, Reyes BE and Labez Carrill OL. Organophorous pesticides exposure alters sperm chromatin structure in mexican agricultural workers. Toxicology and Applied Pharmacology. 2004; 94: 108-13.
29. El-Demerdash FM, Yousef MI, Kedwany FS, Baghdadi HH. Role of alpha-tocopherol beta-carotene in ameliorating fenvalerate induced changes in oxidative stress, hemato-biochemical parameters and semen quality of male rats. Journal of Environmental Science and Health, Part B. 2004; 39: 443-459.
30. Joshi SC, Bansal B, Jasuja ND. Evaluation of reproductive and developmental toxicity of cypermethrin in male albino rats. Toxicology and environmental chemistry. 2011; 93: 592-602.
31. Elbetieha A, Da'as SI, Khamas W and Darmani H. Evaluation the toxic potential of cypermethrin pesticide on some reproductive and fertility parameters in the male rats. Archives of Environmental Contamination and Toxicology. 2001; 41: 522-528.
32. Yang JC, Wu GF, Feng Y, Sum CM. and Hu JM. CSD mRNA expression in rats testes and the effect of taurine on testosterone secretion. Amino acids. 2010; 39: 155-160.
33. Gonzales G, Villena A. True corrected seminal fructose level: a better marker of the function of seminal vesicles in infertile men. International Journal of Andrology. 2001; 24: 255-260.
34. Gupta RS, Sharma R, Sharma A, Bhatnager AK, Dobhal MP, Joshi YC, Sharma MC. Effect of Alstonia scholaris bark extract on testicular function of Wistar rats. Asian Journal of Andrology. 2002; 4: 175-178.

35. Linder RE, Rehnberg GL, Strader LF, Diggs JP. Evaluation of reproductive parameters in adult male Wistar rats after subchronic exposure (gavage) to benomyl. Journal of Toxicology and Environmental Health. 1988; 25: 285-9.
36. Bepari M, Maity P, Das T and Maiti CS. Zinc and αlpha-lipoic acid alleviate cypermethrin induced reproductive toxicity in mature male Wistar rat. International journal of life science and pharma research. 2014; 4: 9-20.
37. Gupta A, Nigam D, Gupta A, Shukla GS and Agarwal AK. Effect of pyrethroid based liquid mosquits repellent inhalation on the blood-brain barrier function and oxidative damage in selected organs of developing rats. Journal of Applied Toxicology. 1999; 19: 67-72.
38. Hayes JD, Flanagan JU, Jowsey IR. Glutathione transferases. Annual Review of Pharmacology and Toxicology. 2005; 45: 51-88.
39. Yamamoto Y, Yamashita S. Plasma ubiquinone to ubiquinol ratio in patients with hepatitis, cirrhosis and hepatoma and in patients treated with percutaneous transluminal coronary reperfusion. Bio Factors. 1999; 9: 241-246.