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Determination of MTD and effect of combined subacute exposure of cypermethrin and deltamethrin and its amelioration by *Withania somnifera* and resveratrol in rats

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Abstract
This experiment was conducted to evaluate the ameliorating effect of *Withania somnifera* and resveratrol against induced changes in body weight gain, relative body weight gain and relative organ weight by combined exposure of cypermethrin and deltamethrin in rats during month of June, 2017.The Cypermethrin and deltamethrin are synthetic pyrethroid insecticides used for pest control in agriculture and as acaricide in animals. The MTD of cypermethrin and deltamethrin in adult male Wistar rats was determined to be 750 mg/kg and 40 mg/kg orally in a pilot dose range finding study. Body weight of each rat was recorded on alternate day till completion of experiment and changes in weight of various body organs (liver, kidney and brain) were examined after sacrifice. Combined treatment of cypermethrin and deltamethrin resulted in decreased body weight gain (both absolute and relative) as compared to naive. A significant change was also observed in relative organ weight in combined cypermethrin and deltamethrin treated groups as compared to naïve which was restored by *Withania somnifera* and resveratrol co-treatment. The study revealed a mild to moderate toxic effect of combined treatment of cypermethrin and deltamethrin on body weight gain and relative organ weight and these toxic effects was ameliorated by *Withania somnifera* and resveratrol.

Keywords: Cypermethrin, Deltamethrin, resveratrol, *Withania somnifera*

1. Introduction
Synthetic pyrethroids is an unique group of insecticides having pyrethrin like structure with better performance characterized and account for over 30% of insecticide use globally [5]. Pyrethroids are modified derivatives of pyrethrins, natural substance obtained from flowers of *Pyrethrum* species. Pyrethroids are widely used in agriculture and veterinary applications due to their high bio-efficacy, enhanced stability and comparatively low mammalian toxicity [5, 11].
Cypermethrin is a synthetic pyrethroid insecticide which is used to kill insects especially on cotton. It behaves as a fast-acting neurotoxin in insects [5]. Cypermethrin is used in agriculture to control ectoparasites which infest cattle, sheep and poultry [14]. In veterinary practice, it is effective in controlling ticks on dogs. Synthetic pyrethroids affect axons of neurons of peripheral nervous system and central nervous system. It interacts with transportation system of sodium ions through cellular membrane. This results a delay in closing of sodium channel and prolonged sodium tail current after membrane gets repolarized. Thus cypermethrin acts as neurotoxic for both insects and mammals [5, 19].
Deltamethrin is a synthetic pyrethroid insecticide used in agriculture, home pest control and disease vector control. Deltamethrin, is used in agriculture, home pest control and disease vector control. It is able to pass from a woman's skin through her blood and into breast milk [4]. Neurotoxic mechanisms of deltamethrin include prolonging the opening of voltage sensitive sodium channels and inhibition of voltage gated chloride channels and GABA receptors [22].
Resveratrol (trans-3, 5, 4-trihydroxystilbene), is a potent antioxidant and cytoprotective agent [6]. Resveratrol (3, 5, 4'-trihydroxy-trans-stilbene) is a stilbenoid and originally isolated by Takaoka from the roots of hellebore in 1940. It attracted wider attention in 1992 when its presence in wine was suggested as the explanation for cardio protective effects of wine [9]. The major dietary sources of stilbenes include grapes, wine, soy, peanuts and peanut products [6, 3]. Resveratrol has been found to exert a number of potentially cardio protective effects *in vitro*,

including promotion of vasodilation by enhancing the production of nitric oxide (NO), inhibition of inflammatory enzymes and antidiabetic effects [8].

Withania somnifera is a plant in the Solanaceae or nightshade family. It is used as an herb in ayurvedic medicine. The main chemical constituents are alkaloids and steroidal lactones [14]. *Withania somnifera* possesses anti-inflammatory, antitumor, antistress, antioxidant, immunomodulatory, hemopoetic, and rejuvenating properties [12, 13]. *Withania somnifera* is a well-known and important medicinal plant widely used in several indigenous system of medicine for treatment of various ailments *viz*. asthma, inflammatory disease, bronchitis, ulcer and stomach problems [14]. Major phytoconstituents of this species are steroidal lactones. Pharmacological experiments in a number of *in vitro* and *in vivo* models have demonstrated the ability of W*ithania somnifera* to exhibit anti-inflammatory, antiulcer, antidiabetic, central nervous system depressant and hepatoprotective activities leading support to the rationale behind several of its traditional uses [12].

The present study was conducted to investigate the ameliorating effect of *Withania somnifera* and resveratrol against induced biochemical changes by combined exposure of cypermethrin and deltamethrin.

2 Materials and Methods

This experiment was conducted in month of June, 2017.

2.1 Drugs and chemicals

Cypermethrin and deltamethrin formulations, were purchased from Bayer Crop Science Ltd., India. Resveratrol was procured from Sigma-Aldrich Company. Methanolic extract of *Withania somnifera* was prepared in the departmental laboratory.

2.2 Animals and treatment

A total of 84 adult male Wistar rats weighing 100-120 g were procured from Disease Free Small Animal House (DFSAH), Lala Lajpat Rai University of Veterinary and Animal Sciences (LUVAS), Hisar, and housed in polyacrylic cages in a group of 7 rats per cage in the departmental animal house. Bedding material (rice husk) was regularly changed on alternate days. The animals were provided with feed and water *ad libitum* and maintained at room temperature with a natural light-dark cycle. Rats were acclimatized to laboratory conditions for 7 days before start of the experiment. Animal house temperature varied between 22 to 27 ºC throughout the investigation. The prior approval of institutional animal ethics committee was obtained for the use of experimental animals in this study. Forty two rats were used for 14 days study, while remaining forty two rats were used for 28 days study.

The rats were randomly divided into six groups, each comprising of seven rats. Group 1 was Naive (control) group which received 3% gum acacia suspension orally. Group 2 was administered cypermethrin (75mg/kg) plus deltamethrin (4mg/kg) as suspension in 3% gum acacia orally. Group 3 animals received cypermethrin (75mg/kg) plus deltamethrin (4mg/kg) as suspension in 3% gum acacia and separately *Withania somnifera* (12.5mg/kg) suspension in 3% gum acacia orally. Group 4 animals were administered cypermethrin (75mg/kg) plus deltamethrin (4 mg/kg) as suspension in 3% gum acacia and separately resveratrol (5 mg/kg) suspension in 3% gum acacia orally. In, Group 5 *Withania somnifera* (12.5 mg/kg) in 3% gum acacia suspension was administered orally, and in Group 6, Resveratrol (5 mg/kg) in 3% gum acacia suspension was administered orally. Experimental groups were same for 14 days as well as for 28 days study.

2.3 Determination of the maximum tolerated dose (MTD):

MTD of cypermethrin and deltamethrin was determined in male rats by oral route following the method as described by Moser and Padilla [16]. The pilot dose range finding studies were conducted in small groups of rats (n=3) using several doses including few lethal doses. Single dose of cypermethrin and deltamethrin was administered in a group of 2-3 animals and observations were made at various time intervals. Thereafter, several iterations were conducted to determine MTD. Out of these doses, a maximum dose was selected that produced clear signs of toxicity but does not result in lethality i.e. maximum tolerated dose (MTD).

2.3.1 Absolute weight gain (g) = final body weight (g) – initial body weight (g)

2.3.2 Relative organ weight (g % or g/100g) = organ weight / body weight X 100

2.3.4 Relative body weight (g % or g/100g) = final body weight – initial body weight / initial body weight X 100

2.4 Statistical Analysis

Data were expressed as mean ± SE. Statistical analysis of data was performed using Graph Pad prism 5.03 and Microsoft Excel. Data were analyzed by ANOVA along with Bonferroni multiple comparison post hoc test. A value of $p < 0.05$ was considered statistically significant.

3. Results

The dose range values and corresponding mortality data in determining the MTD of cypermethrin and deltamethrin in adult male rats by conducting pilot dose range finding study are presented in Table 1 and 2. The MTD of cypermethrin in adult male Wistar rats was found to be 750 mg/kg by oral route and the MTD of deltamethrin in adult male rats was found to be 40 mg/kg by oral route.

Effect of various doses used for determination of MTD on gross observable behaviour were noted in which toxic symptoms started in 10-15 min after cypermthrin and deltamethrin administration and were found to be dose dependent on onset and severity of effect. The main clinical signs found in cypermethrin toxicity were decreased feed intake, loss of body weight, rough hair coat, diarrhoea dyspnoea, ataxia, eye discharge, salivation, increased startle response, somnolence, gradual development of hind limb extensor tone, seizures, incoordination and tremors. The main clinical signs found in deltamethrin toxicity were CNS excitation, which was followed by depression. A variable sequence of motor symptoms developed that involved occasional pawing, or burrowing, coarse whole body tremor associated with movement, gradual development of hind limb extensor tone, chewing, licking and salivation. Finally, choreoathetosis (sinous writhing) developed and animals exhibited slow twisting or writhing movement of neck and tail. At the terminal stage, animals showed laboured breathing, gasping and death. Following combined treatment of cypermethrin and deltamethrin and *Withania somnifera* and resveratrol, body weights of rats were recorded on alternate day till completion of experiment.

3.1 Effect of combined treatment of cypermethrin and deltamethrin on absolute body weight gain (g) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 and 28 days study

Effect of combined treatment of cypermethrin and deltamethrin on absolute body weight gain (g) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 and 28 days study is presented in table 3. Combined treatment of cypermethrin and deltamethrin significantly ($p<0.05$) reduced the absolute body weight gain as compared to naive group of animals. *Withania somnifera* co- treatment along with combined treatment of cypermethrin and deltamethrin significantly ($p<0.05$) restored the absolute body weight gain as compared to combined treatment of cypermethrin and deltamethrin. Resveratrol co-treatment along with combined treatment of cypermethrin and deltamethrin significantly ($p<0.05$) restored the absolute body weight gain as compared to combined treatment of cypermethrin and deltamethrin. *Withania somnifera* treatment alone caused non-significant increase in 14 days study and significantly ($p<0.05$) increase in 28 days study in absolute body weight gain as compared to naive group of animals. Resveratrol treatment alone also caused non-significantly ($p<0.05$) decrease in absolute body weight gain as compared to naive group of animals in 28 days study. Resveratrol treatment alone also caused significantly ($p<0.05$) decrease in absolute body weight gain as compared to *Withania somnifera* treatment alone group in 28 days study.

3.2 Effect of combined treatment of cypermethrin and deltamethrin on relative body weight gain (g/ 100g body wt.) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days and 28 days study

Effect of combined treatment of cypermethrin and deltamethrin on relative body weight gain (g/ 100g body wt.) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days and 28 days study is presented in table 4. Combined treatment of cypermethrin and deltamethrin significantly ($p<0.05$) reduced the relative body weight gain as compared to naive group of animals in both 14 and 28 days study. *Withania somnifera* co-treatment along with combined treatment of cypermethrin and deltamethrin non-significantly ($p<0.05$) restored the relative body weight gain in both 14 and 28 days study. Resveratrol co-treatment along with combined treatment of cypermethrin and deltamethrin non-significantly restored the relative body weight gainin both 14 and 28 days study. *Withania somnifera* treatment alone also caused non-significant ($p<0.05$) increase in relative body weight gain as compared to naive group of animals in both 14 and 28 days study and resveratrol treatment alone also caused non-significant ($p<0.05$) decrease in relative body weight gain as compared to naive groupin both 14 and 28 days study.

3.3 Effect of combined treatment of cypermethrin and deltamethrin on relative organ weight gain (g/100g body wt.) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days and 28 days study

Effect of combined treatment of cypermethrin and deltamethrin on relative organ weight gain (g/100g body wt.) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days and 28 days study ispresented in table 5 and table 6 respectively.No statistically significant changes ($p<0.05$) were observed in liver in 14 days study while combined treatment of cypermethrin and deltamethrin caused significantly ($p<0.05$) increase in relative liver weight as compared to naive group in 28 days study. Both *Withania somnifera* co-treatment and resveratrol co-treatment along with combined treatment of cypermethrin and deltamethrin non-significantly ($p<0.05$) restored the relative liver weight as compared to combined treatment of cypermethrin and deltamethrin in 28 days study.No statistically significant changes ($p<0.05$) were observed in kidney in combined treatment group of cypermethrin and deltamethrin as compared to naive group in both 14 and 28 days study but there was significant changes ($p<0.05$) in brain of combined treatment group of cypermethrin and deltamethrin as compared to naive groupin both 14 and 28 days study. Both *Withania somnifera* co-treatment and resveratrol co-treatment along with combined treatment of cypermethrin and deltamethrin non-significantly ($p<0.05$) restored the relative brain weight in both 14 and 28 days study. *Withania somnifera* and resveratrol treatment alone did not have any effect on the relative brain weight as compared to naive group in both 14 and 28 days study.

Table 1: Maximum tolerated dose (MTD) of cypermethrin administered orally in adult male Wistar rats

Dose (mg/kg)	Number of rats died/ number of rats administered	Percent mortality
800	3/3	100
790	3/3	100
780	2/3	66.66
770	1/3	33.33
760	1/3	33.33
750	0/3	00
740	0/3	00
730	0/3	00

MTD of cypermethrin: 750 mg/kg b.wt. orally

Table 2: Maximum tolerated dose (MTD) of deltamethrin administered orally in adult male Wistar rats

Dose (mg/kg)	Number of rats died/ number of rats administered	Percent mortality
65	3/3	100
60	3/3	100
55	2/3	66.66
50	2/3	66.66
45	1/3	33.33
40	0/3	00
35	0/3	00
30	0/3	00

MTD of deltamethrin: 40 mg/kg b.wt. orally

Table 3: Effect of combined treatment of cypermethrin and deltamethrin on absolute body weight gain (g) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days and 28 days study

Treatment	14 days	28 days
Naïve	29.33 ± 3.26	57.00 ± 2.10
C + D	$10.71^{a} \pm 1.17$	$16.14^{a} \pm 1.18$
C + D + W	$26.87^{b} \pm 2.56$	$47.57^{ab} \pm 1.46$
C + D + R	$25.43^{b} \pm 1.27$	$45.57^{ab} \pm 1.02$
W	$36.86^{bcd} \pm 2.21$	$65.00^{abcd} \pm 1.19$
R	$29.86^{b} \pm 2.25$	$55^{bcde} \pm 1.04$

Values are expressed as Mean ±SEM of seven animals in each group.

a, b, c, d, e ($p \leq 0.05$) vs. control, C + D, C + D + W, C + D + R, W and R, respectively.

C + D mean 10% of MTD of cypermethrin and deltamethrin individually used in combination.

Table 4: Effect of combined treatment of cypermethrin and deltamethrin on relative body weight gain (g/100g body wt.) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 and 28 days study

Treatment	14 days study	28 days study
Naïve	34.31 ± 5.69	49.92± 1.77
C + D	7.39^{a} ± 1.07	13.11^{a} ± 1.12
C + D + W	21.12± 2.84	45.46^{b} ± 1.70
C + D + R	21.86 ± 2.69	44.14^{b} ± 1.50
W	36.49^{b} ± 3.25	55.39^{bcd} ± 1.89
R	26.71^{b} ± 4.40	47.44^{be} ± 2.10

Values are expressed as Mean ±SEM of seven animals in each group.

a, b, c, d, e ($p \leq 0.05$) vs. control, C + D, C + D + W, C + D + R, W and R, respectively.

C + D mean 10% of MTD of cypermethrin and deltamethrin individually used in combination.

Table 5: Effect of combined treatment of cypermethrin and deltamethrin on relative organ weight gain (g/100g) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days study

Treatment	Organs		
	Liver	Kidney	Brain
Naïve	3.20 ± 0.13	0.64 ± 0.10	0.79 ± 0.05
C + D	3.96 ± 0.29	0.77 ± 0.06	0.39a ± 0.07
C + D + W	3.32 ± 0.22	0.66 ± 0.05	0.60 ± 0.06
C + D + R	3.36 ± 0.16	0.67 ± 0.07	0.61 ± 0.03
W	3.15 ± 0.12	0.63 ± 0.13	0.79b ± 0.08
R	3.19 ± 0.42	0.62 ± 0.05	0.81b ± 0.06

Values are expressed as Mean ±SEM of seven animals in each group.

a, b, c, d, e ($p \leq 0.05$) vs. control, C + D, C + D + W, C + D + R, W and R, respectively.

C + D mean 10% of MTD of cypermethrin and deltamethrin individually used in combination.

Table 6: Effect of combined treatment of cypermethrin and deltamethrin on relative organ weight gain (g/100g b.wt.) and its amelioration by *Withania somnifera*and resveratrol in adult male rats in 28 days study

Treatment	Organs		
	Liver	Kidney	Brain
Naïve	3.55± 0.13	0.70± 0.09	0.95± 0.12
C + D	5.82^{a} ± 1.11	1.28± 0.29	0.37^{a} ± 0.04
C + D + W	4.24± 0.14	0.86± 0.09	0.85± 0.17
C + D + R	4.41± 0.29	0.90± 0.12	0.88± 0.11
W	3.51^{b} ± 0.13	0.70± 0.06	0.92^{b} ± 0.10
R	3.59^{b} ± 0.21	0.73± 0.11	0.99^{b} ± 0.13

Values are expressed as Mean ±SEM of seven animals in each group.

a, b, c, d, e ($p \leq 0.05$) vs. control, C + D, C + D + W, C + D + R, W and R, respectively.

C + D mean 10% of MTD of cypermethrin and deltamethrin individually used in combination.

4 Discussion

4.1 Maximum Tolerated Dose (MTD)

MTD of cypermethrin and deltamethrin was evaluated by dose range finding studies in adult male Wistar rats following oral administration. The MTD was found to be 750 mg /kg for cypermethrin and 40 mg /kg for deltamethrin.

4.2 Effect of combined treatment of cypermethrin and deltamethrin on body weight gain and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days as well as in 28 days study

The weight gain in animals serves as index of growth rate [17]. It is obvious that monitoring of body weight provides information on general health level of animals which can also be an important interpretation of reproductive effects [2]. A significant reduction in both absolute (g) and relative body weight gain (g/100 g) was observed in the present study in male rats with combined exposure of cypermethrin and deltamethrin in 14 days as well as in 28 days study. It may be due to decreased food intake. Alwan and Yassin [1] also reported decrease in body weight on oral administration of cypermethrin to rabbits. Mongi *et al.* [15] observed a reduction in weight gain in deltamethrin treated group in rats. The reduction in body weight is probably attributed to the reduction in food intake and toxic effects of cypermethrin and deltamethrin in treated rats. The decrease of weight gain in deltamethrin-treated animals was not simply a reflection of decreased food consumption, but rather due to toxicity of cypermethrin and deltamethrin, perhaps by malabsorption of nutrients induced by effects on the gastro-intestinal tract or inhibition of protein synthesis.

Resveratrol co-treatment and *Withania somnifera* co-treatment along with combined treatment of cypermethrin and deltamethrin significantly restored the absolute body weight gain in 14 days as well as in 28 days study. This may be due to the hepato-protective effect of resveratrol or by reduction in protein catabolism caused by combined treatment of cypermethrin and deltamethrin. *Withania somnifera* alone treatment also significantly increased the absolute body weight gain in 28 days study. This might be due to its properties like antistress, immunomodulatory, haemopoietic properties and its effects on endocrine and cardiopulmonary system [14].

Resveratrol co-treatment and *Withania somnifera* co-treatment alongwith combined treatment of cypermethrin and deltamethrin non-significantly restored the relative body weight gain in 14 days study and significantly in 28 days study.

4.3 Effect of combined treatment of cypermethrin and deltamethrinon relative organ weight gain (g/100g) and its amelioration by *Withania somnifera* and resveratrol in adult male rats in 14 days as well as in 28 days study

In toxicological studies, relative organ weights are important criteria for evaluation of organ toxicity [7, 20].

Liver: In 14 days study, no statistically significant change was observed in relative organ weight of liver. Our results are in accordance with Yavasolgu *et al.* [21] who also reported no significant change in relative organ weight of liver on oral administration of cypermethrin in rats. A statistically significant increase was observed in relative weight of liver in combined cypermethrin and deltamethrin treated rats in 28 days study. The results are in agreement with Grewal *et al.* [10] where a significant increase in relative liver weight was found on oral administration of cypermethrin in rats. Similar results were also recorded by Sharma *et al.* [18] on orally administered deltamethrin in rats. Liver is the main organ of cypermethrin and deltamethrin metabolism. Thus, physiologically the liver could be affected directly by combined treatment of cypermethrin and deltamethrin during the period of exposure in rats.

In present study, resveratrol co-treatment and *Withania somnifera* co-treatment non-significantly reduced the increased relative liver weight by combined exposure of cypermethrin and deltamethrin for 28 days in rats.

4.3.1 Kidney: No statistically significant changes were observed in relative organ weight of kidney in 14 days as well as in 28 days study.

4.3.2 Brain: In present study, significant changes were observed in the relative weight of brain in 14 days as well as in 28 days study. Combined treatment of cypermethrin and deltamethrin significantly caused decrease in brain weight as compared to naive group in 14 days as well as in 28 days study. Cypermethrin and deltamethrin accumulation may cause brain damage which may be responsible for decrease in weight of brain. These results are similar as recorded by Grewal *et al.* [10] and Sharma *et al.* [18] on oral administration of cypermethrin in rats.

In present study, resveratrol co-treatment and *Withania somnifera* co-treatment non-significantly increased the reduced relative brain weight by combined exposure of cypermethrin and deltamethrin for 14 days as well as for 28 days treatment in rats.

5. Conclusion

Combined treatment of cypermethrin and deltamethrin significantly decreased absolute body weight gain and relative body weight gain which was significantly increased by resveratrol co-treatment and *Withania somnifera* co-treatment in 14 as well as in 28 days study. No significant change in relative liver weight was observed by combined treatment of cypermethrin and deltamethrin in 14 days study, while relative liver weight significantly increased in 28 days study. Resveratrol co-treatment and *Withania somnifera* co-treatment non-significantly reduced the increased relative liver weight in both 14 as well as in 28 days study. No significant change in relative kidney weight was observed in between various treatment groups in 14 as well as in 28 days study. Relative brain weight was significantly decreased in combined cypermethrin and deltamethrin exposed rats in both 14 as well as in 28 days study. Resveratrol co-treatment and *Withania somnifera* co-treatment non-significantly increased relative brain weight in both 14 as well as in 28 days study.

5.1 Future Prospective

These two insecticides are used indiscriminately in crops by the farmers to combat insects and leads to toxicity in animals. Farmers use these insecticides in fodder crops and are not aware about the toxic effects on their animals. Sometimes these insecticides also lead the animals to death. Resveratrol co-treatment and *Withania somnifera* co-treatment may help to save the lives of these animals. Present findings may also help the field veterinarians to use these treatments to retard toxicity of these insecticides. More avenues are also there to study about the anti toxic properties of resveratrol and *Withania somnifera* against other groups of insecticides.

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7. References

1. Alwan AK, Yassin MM. Toxic effects of cypermethrin on liver and kidney of male domestic rabbits. Master of Science thesis, 2015, 1-90.
2. Aly H, Domenech O, Ashraf B, Abdel N. Aroclor impairs spermatogenesis and induces oxidative stress in rat testicular mitochondria. Food Chemistry and Toxicology. 2009; 47(8):1733-38.
3. Baur JA, Pearson KJ, Price NL, Jamieson HA, Lerin C *et al*. Resveratrol improves health and survival of mice on a high-calorie diet. Nature. 2006; 444(7117):337-342.
4. Bouwman H, Sereda B, Meinhardt HM. Simultaneous presence of DDT and pyrethroid residues in human breast milk from a malaria endemic area in South Africa. Environmental pollution. 2006; 144(3):902-917.
5. Casida JE, Gammon DW, Glickman AH, Lawrence LJ. Mechanisms of selective action of pyrethroid insecticides. Annual review of Pharmacology and Toxicology. 1983; 23**(**1):413-438.
6. Cassidy A, Hanley B, Lamuela-Ravento RM. Isoflavone, lignans and stilbenes-origins, metabolism and potential importance to human health. Journal Science of Food Agriculture. 2000; 80:1044-1062.
7. Crissman JW, Goodman DG, Hildebrandt PK, Maronpot RR, Prater DA, Riley JH. Best practice guideline. Toxicology and Pathology. 2004; 32:126-31.
8. Duffy S, Vita J, Joseph A. Effects of phenolics on vascular endothelial function. Current Opinion in Lipidology. 2003; 14(1):21-27.
9. Fremont L. Biological effects of resveratrol. Life Science. 2000; 66(8):663-673.
10. Grewal KK, Sandhu GS, Kaur RR, Brar RS, Sandhu HS. Toxic impact of cypermethrin onbehaviour and histology of certain tissues of albino rats. International Journal of Toxicology. 2010; 17:94-98.
11. Kale M, Rathore N, John S, Bhatnagar D. Lipid peroxidative damage on pyrethroid exposure and alterations in antioxidant status in rat erythrocytes: a possible involvement of reactive oxygen species. Toxicology letters. 2010; 105(3):197-205.
12. Maheswari R, Manisha P. Withania Somnifera L root extract ameliorates toxin induced cytotoxicity. International Journal of Pharma Sciences and Research. 2015; 6(5):848-855.
13. Mirjalili MH, Moyano E, Bonfill M, Cusido RM, Palazon J. Steroidal lactones from Withania somnifera, an ancient plant for novel medicine. Molecules. 2009; 14(7):2373-2393.
14. Mishra LC, Singh BB, Dagenais S. Scientific basis for the therapeutic use of Withania somnifera (ashwagandha): a review. Alternative medicine review. 2000; 5(4):334-346.
15. Mongi S, Mahfoud M, Amel B, Kamel, Abdelfattah EI. Protective effects of vitamin C against haematological and biochemical toxicity induced by deltamethrin in male Wistar rats. Ecotoxicolgy and Environmental Safety. 2011; 74(6):1765-1769.
16. Moser VC, Padilla S. Age and gender related differences in the time course of behavioural and biochemical effects produced by oral chlorpyrifos in rats. Toxicology and Applied Pharmacology. 1997; 49:161-170.
17. Palani V, Senthikumaran RK, Govindawamy S. Biochemical evaluation of antitumor effect of Muthumarunthu (herbal formulation) on experimental fibrosarcoma in rats. Ethnopharmacology. 1999; 65:257-65.
18. Sharma P, Singh R, Jan M. Dose-dependent effect of deltamethrin in testis, liver, and kidney of Wistar rats. Toxicology International. 2014; 21(2):131.

19. Somasani A. Management of tick infestation in dogs. Journal of Advanced Veterinary and Animal Research. 2014; 1(3):145-147.
20. Timbrell JA. Biomarkers of organ toxicity. Archives of Industrial Hygiene and Toxicology. 2000; 33:295-02.
21. Yavasoglu A, Sayim F, Uyanikgil Y, Turgut M, Karabay-Yavasoglu NU. The pyrethroid cypermethrin-induced biochemical and histological alterations in rat liver. Journal of Health Science. 2006; 52(6):774-780.
22. Yousef MI, Awad TI, Mohamed EH. Deltamethrin-induced oxidative damage and biochemical alterations in rat and its attenuation by Vitamin E. Toxicology. 2006; 227(3):240-247.