International Journal of Immunology Research
ISSN: 0976-4909 & E-ISSN: 0976-4917, Volume 3, Issue 1, 2013, pp.-21-24.
Available online at http://www.bioinfopublication.org/jouarchive.php?opt=&jouid=BPJ0000229

MODULATORY EFFECT OF VITAMIN E AGAINST FENVALERATE INDUCED IMMUNOTOXICITY IN ALBINO MICE

LAMFON H.A.*

Department of Biology, Faculty of Applied Sciences, Umm Al-Qura University, Makkah, Saudi Arabia.
*Corresponding Author: Email- halamf@uqu.edu.sa

Received: July 23, 2013; Accepted: August 03, 2013

Abstract- Fenvalerate is a synthetic pyrethroid commonly used in agriculture and other domestic applications. The present work studied the effect of vitamin E on immunotoxicity of fenvalerate in albino rats. Treating animals with fenvalerate (1/10 LD_{50}, 3 times weekly for 2 weeks) caused obvious histopathological alterations in the thymus represented by depletion of cortical thymocytes, atrophy of medulla, hemorrhages in the stroma and focal areas of necrosis. Hematological results showed an increase in the total leucocytes, in the percentage of lymphocytes and a significant decrease in percentages of monocytes and neutrophilis. Biochemical results revealed an increase in MDA and a decrease in the activity of SOD and CAT. Treating mice with fenvalerate and vitamin E (20mg/kg body weight) improved the architecture of the thymus. The thymus showed an increase in the cellularity and the number of leukocytes becomes nearly to the normal value. Moreover, the activity of SOD and CAT was enhanced and lipid peroxidation was diminished. These effects are attributed to the antioxidant activity of vitamin E.

Keywords- Fenvalerate, Thymus, Vitamin E, Mice, Antioxidant

Citation: Lamfon H.A. (2013) Modulatory Effect of Vitamin E against Fenvalerate Induced Immunotoxicity in Albino Mice. International Journal of Immunology Research, ISSN: 0976-4909 & E-ISSN: 0976-4917, Volume 3, Issue 1, pp.-21-24.

Introduction

Pyrethroids are one of the most widely used insecticides. The studies conducted on animals indicate that the toxicity of pyrethroids depends on many factors, such as body construction, route of administration and period of administration. The source of pyrethroids is the flowers of the pyretherum plant *Chrysanthemum cinerariafolium* [1]. Due to the persistence of these insecticides in the environment, structures similar to pyrethroids have been synthesized and proved to be effective against different insects [2]. Fenvalerate is a synthetic pyrethroid commonly used in agriculture and other domestic applications due to its high insecticidal activity and low mammalian toxicity. Its widespread use has however been associated with adverse consequences on animal health [3]. High doses of fenvalerate has been reported to cause reduction of body mass, increase in liver mass, and proliferation of the smooth endoplasmic reticulum in hepatic cells, and induction of the activity of microsomal enzymes [4,5].

Vitamin E is one of the most important lipid-soluble antioxidants occur in plants and animals, specifically for protection against lipid peroxidation in biological membranes. Vitamin E reduces oxidative stress, improves vascular function and structure, and prevents progression of hypertension in adult stroke-prone spontaneously hypertensive rats [6]. Minko, et al., [7] examined the antioxidant and antiapoptotic activity of vitamin E rats exposed to severe hypoxia. They showed that intratracheal application of vitamin E normalized lung phospholipid composition and inhibited lipid peroxidation in lung tissues, which in turn decreased lung edema damage, improved breathing pattern, Oxygen diffusion and lung gas exchange. Latchoumycandane and Mathur [8] reported that vitamin E protect against 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) induced oxidative stress in the rat testis. The present study was designed to determine the effect of vitamin E on immunotoxicity of pyrethroid insecticide, fenvalerate in albino rats.

Materials and methods

Experimental animals

Healthy adult male albino mice (*Mus musculus*) approximately 3 months old weighing 20 ± 5g were used in the present study. Animals were kept in the laboratory under constant condition of temperature (24 ± 2 °C) for at least one week before and throughout the experimental work. They were maintained on a standard rodent diet. Water was available ad libitum. All the experiments were done in compliance with the guide for the care and use of laboratory animals [9]. Animals were divided into four groups, 20 animals in each group. The first group served as a control, while animals of the second group were given vitamin E at a dose level of 20mg/kg body weight orally three times/ week for 2 weeks. Animals of the third group were given fenvalerate at a dose level of 1/10 LD_{50} (10 mg/kg body weight) orally by stomach tube three times/week for 2 weeks. Animals of the fourth group were given fenvalerate (1/10 LD_{50}) and vitamin E (20mg/kg body weight) orally at the same time for 2 weeks. Half of the animals from each group were killed by cervical decapitation after one week and the rest of animals were killed after 2 weeks.

Histological studies

Animals were dissected and their thymuses were removed. For histological preparations, the thymus was fixed in alcoholic Bouin's fluid, dehydrated, cleared and embedded in paraffin wax. Paraffin sections of 5 micron thickness were prepared and stained with Ehrlich's haematoxylin and eosin.

Hematological and Biochemical Studies

Blood was collected from control and treated animals after 1 and 2 weeks of treatment. Total leukocytes count and differential counts were measured by a fully automated Coulter counter (Coulter Electronics Limited, England). For biochemical study sera were obtained by centrifugation of the blood samples and stored at 20°C until assayed for the biochemical parameters. The extent of lipid peroxidation was estimated as the concentration of thiobarbituric acid reactive products (malondialdehyde, MDA) according to Ohkawa, et al. [10]. The activity of superoxide dismutase (SOD) was measured using the methods of Rest and Spitznagel [11]. Catalase (CAT) activity was determined from the rate of decomposition of H_2O_2 [12].

Statistical Analysis

Data were expressed as mean values ± SD and statistical analysis was performed using one way ANOVA to assess significant differences among treatment groups. The criterion for statistical significance was set at $p<0.05$. All statistical analyses were performed using SPSS statistical version 16 software package (SPSS® 4 Inc., USA).

Results

Histological Observations of Thymus

The thymus is a large flattened and bilobed lymphoid organ lies over the heart. Each lobe is surrounded by a connective tissue capsule which extends inward deep to give trabeculae dividing each lobe into many pseudo-lobules. Histologically, the thymus consists of several discrete area: the connective tissue capsule which surrounds the cortex, which itself surrounds the medulla [Fig-1a]. The cortex contains thymocytes in addition of macrophages. The medulla has abundant epithelial reticular cells, thymocytes and lymphocytes of medium and large size. Treating animals with fenvalerate for 7 days revealed obvious histopathological alterations represented by absence of cortico-medullary demarcation and appearance of degenerated areas characterized by necrotic cells with pyknotic nuclei [Fig-1b]. An increase in these histopathological changes was observed after 14 days. The cortical thymocytes were depleted and the medulla atrophied. Hemorrhages in the stroma and focal areas of necrosis were observed [Fig-2a]. Marked amendment in thymic architecture was recorded in animals given fenvalerate and vitamin-E. This was particularly ratified as increase in the density of cortical thymocytes and appearance of normal trabeculae [Fig-2b]. Data in [Table-1] showed the change in cortical thickness in different animal groups.

Change in Leukocytes Counts

Data in [Table-2] showed that treating mice with fenvalerate caused an increase in mean number of leukocytes compared with controls. Co-administration of vitamin-c restored the number of leukocytes nearly to the normal value. Treating animals with fenvalerate caused an increase in the percentage of lymphocytes and a significant decrease in percentages of monocytes and neutrophilis. On the other hand, treating animals with fenvalerate and vitamin E showed normal values of these parameters.

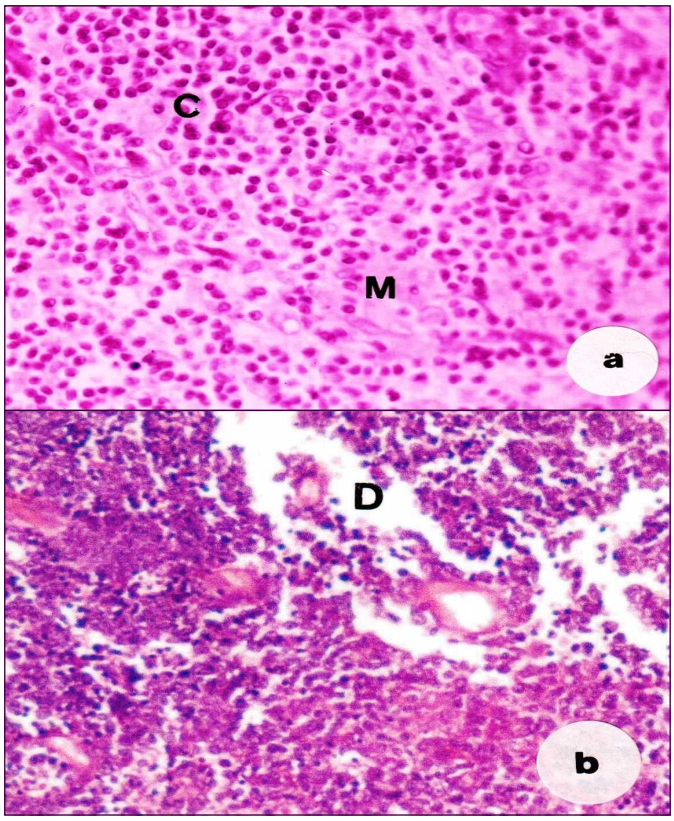

Fig. 1- (a) Section in the thymus of a control mouse showing cortex (C) and medulla (M), **(b)-** Thymus of a mouse treated with fenvalerate for one week showing degenerated area (D), X300.

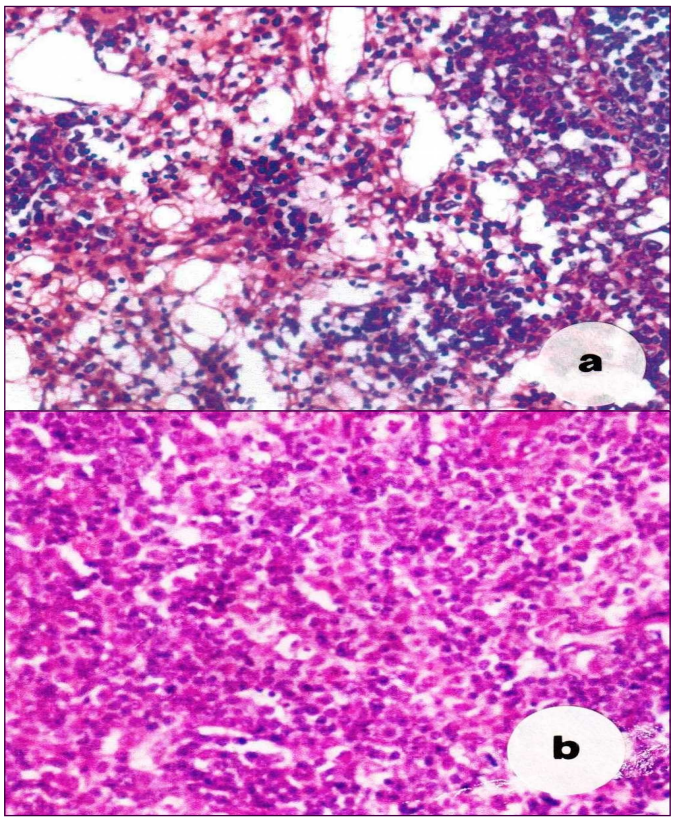

Fig. 2- (a) Atrophied thymus of a mouse treated with fenvalerate for 2 weeks. **(b)-** Thymus of a mouse treated with fenvalerate and vitamin E showing increase of cellularity, X300.

Table 1- The mean cortical thickness of the thymus gland after different treatments

Animal Group	Cortical thickness (μ) Mean + SD
Control group	119 ± 13
Vitamin E	117 ± 11
Fenvalerate	74 ± 7*
Fenvalerate+ Vit.E	98 ± 9

(*).Significant at p<0.05

Table 2- Change in percentages of lymphocytes, monocytes and neutrophils in different animal groups after 2 weeks

Animal group	Leukocytes (10^6/mm^3)	Lymphocyte (%)	Monocytes (%)	Neutrophils (%)
Control	4.5 ± 0.9	51.9 ± 4.2	8.3 ± 1.2	22.9 ± 3.2
Vit. E	4.9 ± 0.4	50.5 ± 1.8	8.5 ± 1.1	23.5 ± 2.3
Fenvalerate	6.8 ± 1.2*	56.6 ± 6.1*	2.7 ± 0.9*	16.6 ± 1.4*
Fenvalerate+Vit.E	4.9 ± 1.1	51.2 ± 5	5.4 ± 1.3	19.5 ± 1.1

(*).Significant at p<0.05

Biochemical Results

Data in [Table-3] revealed significant (p<0.05) increase in the level of MDA in sera of animals treated with fenvalerate compared with the control group. Animals treated with fenvalerate and vitamin E showed decrease in MDA levels in sera compared with fenvalerate group. A decrease in the activity of SOD and CAT was recorded in sera of mice exposed to fenvalerate compared with control group. Animals given fenvalerate and vitamin E showed significant increase in the activity of SOD and CAT compared with fenvalerate group.

Table 3- Change in MDA, CAT and SOD in different animal groups after 2 weeks

Animal group	MDA (nmol/ml)	CAT (umol/sec/ml)	SOD (nmol/ml)
Control	28 ± 1.8	1.45 ± 0.02	52.4 ± 2.5
Vit. E	29 ± 1.1	1.50 ± 0.01	54.2 ± 1.9
Fenvalerate	39 ± 2.1*	0.40 ± 0.01*	22.3 ± 2.1*
Fenvalerate+Vit.E	31 ± 1.6	1.10 ± 0.1	40.5 ± 3.2

(*).Significant at p<0.05

Discussion

Thymus gland is essential for life as it controls lymphopoiesis in central and peripheral lymphoid organs and differentiation of lymphocytes into immunologically competent T-cell [13]. The present results revealed that fenvalerate affected the thymus of mice. Several authors reported impairment of haematopoietic system, including thymus, following exposure to insecticides. Madsen, et al., [14] reported that oral administration of rats with deltamethrin and alpha-cypermethrin increased weight of mesenterial lymph nodes, decreased thymus weight and increased antibody production and splenic natural killer-cell activity. After an intraperitoneal dose of deltamethrin, thymus atrophy was observed in mice in a dose-dependent manner [15].Topical permethrin application caused 32% inhibition of splenic T cell proliferation; in vitro exposure to permethrin also diminished splenocyte proliferation. Dose-related decreases in thymic cellularity of 52 and 80% were seen in mice exposed to 15 and 25 microl permethrin, respectively. Apoptosis was significantly increased in CD4(-)8(-) and CD4(-)8(+) thymocytes, and the CD4(+)CD8(+) thymocyte subpopulation was most severely diminished [16]. Morgan and Osman [17] reported that Lambda-cyhalothrin caused leucopenia, lymphopenia, depletion of lymphoid cells in the white pulps of spleen, mesentric lymph nodes and Peyer's patches and severe atrophy of thymus cortex in rabbits.

The present result indicated that the total number of leucocytes was significantly increased in the treated mice. An increase in the percentage of lymphocytes and a significant decrease in percentages of monocytes and neutrophilis were also recorded. Similarly, Haratym-Maj [18] reported that alpha-cypermethrin, deltamethrin and fenvalerate, irrespective of the sex of mice, caused an increase in the number of leukocytes and decrease in the percentage of monocytes and neutrophilis in peripheral blood. This finding was obtained by some investigators who observed hematological changes in mammalian animals treated with pyrethroid insecticides. Sub-acute poisoning with alpha -cypermethrin in female Swiss mice, however, results in an increase in the general number of leukocytes (monocytes and lymphocytes) and does not cause changes in the erythrocytic system [4]. Yousef, et al. [19] reported that cypermethrin significantly (p<0.05) decreased hemoglobin, total erythrocytic count and packed cell volume, while total leukocyte count increased in rabbits.It was suggested that the increase in the leukocytes may result from the mobilization of the immunological system and/or a shift in the leukocytic pool from the spleen to peripheral blood.

Treating animals with fenvalerate decreased the activity levels of SOD and CAT while lipid peroxidation was high as evidenced by increase in the level of MDA. In agreement with these results, El-Demerdash, et al. [20] showed that fenvalerate significantly (p<0.05) induced free radicals in plasma and brain and insignificantly in liver and testes. Prasanthi, et al. [21] reported that fenvalerate and its metabolite have the propensity to cause significant oxidative damage in rat erythrocytes in vitro, which is associated with marked damage to membrane proteins. Fenvalerate exposure in normal rats increased hepatic glutathione peroxidase activity and lipid peroxidation, decreased glutathione content, but did not change the activities of catalase or any of the superoxide dismutase forms [22]. Desi, et al., [23] reported that injury to the immune system may be caused by increased reactive oxygen species (ROS) production. Galloway and Handy [24] suggested that malathion caused toxicity in the immune system via oxidative stress. The authors recommended that immunotoxicity was likely to occur through oxidative damage to immune organs, or via chronic effects of altered metabolism/nutrition on immune organs.

Antioxidants have been reported to provide protection from the toxicity of pesticides. The obtained results showed that coadministration of vitamin E ameliorate the toxicity of fenvalerate. The thymus showed an increase in the cellularity and the number of leukocytes becomes nearly to the normal value. Moreover, the activity of SOD and CAT was enhanced and lipid peroxidation was diminished. Similarly, Alsharif and Hassoun [25] demonstrated that vitamin E succinate resulted in partial protection against TCDD-induced thymic atrophy and oxidative stress in mice. Petrova and Donchenko [26] studied the effect of α-tocopherol on actinomycin D induced apoptosis in rat thymus and they found that the induction of rat thymocyte apoptosis by actinomycin D was associated with the increased caspase-3 activity and DNA fragmentation, both effects were attenuated by α-tocopherol. Minko, et al., [7] examined the antioxidant and antiapoptotic activity of vitamin E in anesthetized rats exposed to severe hypoxia. They shown that intratracheal application of vitamin E normalized lung phospholipid composition and inhibited lipid peroxidation in lung tissues, which in turn decreased lung edema damage, improved breathing pattern, Oxygen diffusion

and lung gas exchange. Vitamin E has been shown to inhibit the free-radical-induced damage to sensitive cell membranes as it is a major chain-breaking antioxidant [27]. It is suggested from the obtained results that vitamin E may directly quench the free radicals such as peroxyl and alkoxyl. Thus, by scavenging these radicals, it breaks free-radical chain reaction and forms a relatively stable complex such as tocopheroxyl radical [28]. Thus, vitamin E protects the thymus and immunosystem against the damages caused by reactive oxygen species produced by fenvalerate

References

[1] Casida J.E. (1973) *Pyrethrum, the Natural Insecticide*, Academic Press, New York.

[2] McEwen F.L. and Stephenson G.R. (1979) *The Use and Significance of Pesticides in the Environment*, John Wiley and Sons, New York.

[3] Hemming H., Flodstrom S., Warngard L. (1993) *Carcinogenesis,* 14(12), 2531-2535.

[4] Luty S.T., Maj A.H., Latuszynska J., Daniela O., Rodak T.M. (2001) *Ann. Agric. Environ. Med.*, 8(2), 245-254.

[5] Mani U., Prasad A.K., Sureshkumar V., Kumar P., Kewallal K. Maji B.K., Dutta K.K. (2004) *Biomed. Environ. Sci.,* 17, 309-314.

[6] Chen X., Touyz R.M., Park J.B., Schiffrin E.L. (2001) *Hypertension,* 38(3), 606-11.

[7] Minko T., Stefanov A., Pozharov V. (2002) *J. Appl. Physiol.*, 93 (4), 1550-1560.

[8] Latchoumycandane C. and Mathur P.P. (2002) *J. Appl. Toxicol.,* 22(5)*,* 345-351.

[9] National Research Council (1985) *Guide for Use and Care of Laboratory Animals*, 85-23, NIH, Washington, DC.

[10] Ohkawa H., Ohishi N., Yagi K. (1979) *Annals of Biochemistry*, 95, 351-358.

[11] Rest R.F. and Spitznagel J.K. (1977) *Biochem. J*., 166, 145-153.

[12] Aebi H., Wyss S.R., Scherze B. and Skvaril F. (1974)*Enzyme*, 17(5), 307-318.

[13] Leeson C.R. and Leeson T.S. (1981) *Histology*, 4th ed., W.B. Saundres Co., London, UK.

[14] Madsen C., Claesson M.H., Ropke C. (1996) *Toxicology*, 107 (3), 219-227.

[15] Enan E., Pinkerton K.E., Peake J., Matsumura F. (1996) *Pharmacol.*, 51(4), 447-454.

[16] Prater M.R., Gogal R.M., Blaylock B.L., Longstreth J., Holladay S.D. (2002) *Food and Chemical Toxicology,* 40(12), 1863-1873.

[17] Morgan A.M. and Osman A.H. (2007) *J. Egypt. Soc. Toxicol.,* 36, 23-33.

[18] Haratym-Maj A. (2002) *Ann. Agric. Environ. Med.*, 9(2), 199-206.

[19] Yousef M.I., El-Demerdash F.M., Kamel K.I., Al-Salhen K.S. (2003) *Toxicology,* 189(3), 223-234.

[20] El-Demerdash F.M., Yousef M.I., Kedwany F.S., Baghdadi H.H. (2004) *J. Environmental Science and Health*, 39(3), 443-459.

[21] Prasanthi K., Rajini P.S. (2005) *Toxicol. In Vitro*, 19(4), 449-456.

[22] Giray B. and Hincal F. (2011) *Hum. Exp. Toxicol.,* 30(10), 1575-83.

[23] Desi I., Dobronyi I., Varga L. (1986) *Ecotoxicol. Environ. Saf.*, 12(3), 220-232.

[24] Galloway T. and Handy R. (2003) *Ecotoxicology,* 12(1-4), 345-363.

[25] Alsharif N.Z. and Hassoun E.A. (2004) *Basic Clini. Pharmacol. Toxicol*., 95(3), 131-8.

[26] Petrova G.V. and Donchenko G.V. (2005) *Ukr. Biokhim Zh*., 77 (1), 72-77.

[27] Sinclair S. (2000) *Alternative Medicine Review,* 5(1), 28-38.

[28] Verma A. and Kanwar K.C. (1999) *Asian Journal of Andrology,* 1(3), 151-154.