

Change in acetylcholine activity and some blood parameters in adult sheep dipped in deltamethrin

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Abstract

The aim of this was to elucidate the effects of deltamethrin dipping on acetylcholine activity in serum and some blood parameters of sheep. The study was conducted on forty adult sheep dipped in deltamethrin in concentration of 1:1000L of water. Venous blood samples were collected from each animal before and after dipped. There was significant inhibition in the acetylcholine esterase in all subjected animals after dipping. Mean values of hematological investigations showed a significant rise in the number of total leucocytes (leukocytosis) with significant decline in hemoglobin concentration and Red cell indices in all dipped animals after dipping in deltamethrin when compared with mean values obtained from the same animals before dipping. Conclusion of our results revealed that poisoning with deltamethrin affects blood parameters through the inhibition of acetylcholine esterase.

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Introduction

Organophosphorus compound and carbamate pesticides are broadly used in public health (1), veterinary practice, and in agriculture (2). They pose main environmental pollution complications and health threats to people and animals (3,4). These pesticides inhibit cholinesterase (ChE) activity in the nervous tissues and neuromuscular junctions, causing an accumulation of acetylcholine at the nerve endings which afterward produces signs of toxicosis characterized by nicotinic, muscarinic, and central nervous system effects (5,6). The Cholinesterase activities in blood, tissue and brain can be easily measured (7,8) and their investigations are useful in determining the exposure status of cholinesterase inhibitors and detecting the toxicity of these compounds (8-10). Acetylcholinesterase activity inhibition have been widely studied red blood cells, plasma and tissues in workers (11,12), experimental animals (13,14), fish (9), birds (15,16) and in farm animals (8,17) after exposure to organophosphate and carbamate pesticides but rarely studied in serum (18) specially in our country and adverse effect on

blood parameters. One of the basic mechanisms of the harmfulness of the pesticides seems to be lipid peroxidation; as a consequence, these composites can lead to some blood cells disorders (19). So this study was designed to elucidate the effects of deltamethrin on acetylcholine and some blood parameters of sheep dipped in deltamethrin.

Material and methods

Sample collection

This study was conducted on forty adult sheep dipped in pesticides. Venous blood samples were collected from each animal; one before dipping (used as control, and the second sample collected after dipping, then each sample was divided into two parts; first part used for complete blood pictures study and the remainder was centrifuged, after clotting, at 2000-3000 rpm for 20 minutes, and the separated serum was immediately frozen at -20°C for later estimation of Ach esterase activity.

Methods

Blood parameters red blood cells (RBC) count, white blood cells (WBC) count, differential leucocyte count, hematocrit, hemoglobin concentration, red cell indices estimated by automated machine (Ruby- automated hematology analyzer) (20). Acetyl choline esterase assayed quantitatively in vitro by using the enzyme linked immunosorbent assay (ELISA) kit (RelAssay Diagnostics) based on the principle of biotin double antibody sandwich technology.

Dipping

Animals were stopped giving water for 12 hours. One hour prior to dipping they were allowed one hour for taking water. Then All subjected animals were dipped in a basin, 0.9*1.5*3 m in dimension, containing deltamethrin 25 g/L (DELTDIP®) in concentration 1:1000L for 30 seconds.

Statistical analysis

Data obtained were analyzed using the Excel program version 10 package. The significance of invariant differences was assessed by student's t- tests and all values were two-sided. A P<0.01 was considered statistically significant.

Results

There was significant decrease in the acetylcholine esterase activity in all subjected animals after dipping as compared to pre-dipping period (Figure 1). The mean values were 5.74 and 1.24±0.31 nmol/Ml before and after dipping respectively.

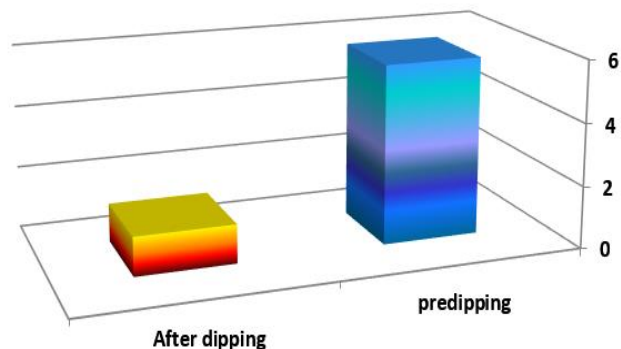


Figure 1: Mean values± SD of Acetylcholine esterase activities in dipped sheep before -and- after dipping in deltamethrin.

Mean values of hematological investigations showed a significant rise in the number of total leucocytes with significant decline in hemoglobin concentration in all animals after dipping in pesticides when compared with mean values obtained from the same animals before dipping (table1). Regarding the differential leucocyte count, our result showed significant increase in the granular (neutrophils and eosinophils) and some granular leucocytes (lymphocytes and monocytes). Mean values of Red cell indices; Mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and Mean corpuscular hemoglobin concentration (MCHC), also showed significant decrease. There was a decrease in the mean value of red blood cells (RBCs) count and hematocrit (Hct) but was statistically no significant.

Table 1: Mean values ± SD of blood parameters in dipped sheep before -and- after dipping in deltamethrin

	WBC 10 ⁶ / m ³	LYM %	MON %	NEU %	EOS %	RBC m/m ³	MCV (fl)	Hct (%)	MCH (pg)	MCHC (g/dL)	HB (g/dL)
Before dipping	13.40 ±1.30	44.06 ±1.06	13.1 ±1.25	23.15 ±0.7	0.26 ±0.03	4.0 ±0.37	32.49 ±0.8	27.86 ±0.83	23.76 ±0.82	80.99 ±1.23	10.3 ±0.38
After dipping	19.7*	62.6*	16.6*	26.6	0.61*	3.77	28.4*	28.13	16.36*	75.8*	8.7*
dipping	±1.8	±1.86	±1.63	±0.93	±0.07	±0.36	±28.46	±0.53	±1.04	±0.71	±0.34

* P<0.01 statistically significant.

Discussion

Organophosphorus compounds (OP) is progressively increasing in Iraq, most likely because of the comprehensive availability of pesticides as a result of wide-ranging use in agriculture, veterinary and also because of over the counter sale of these items. Current results showed a significant decrease in acetylcholine activities in the sera of sheep after dipping in pesticides when compared with the values obtained after dipping. This conclusion confirms the effect of these compounds on acetylcholine activity and put forward that estimation of serum choline esterase is useful in

conformation of OP poisoning. The reported results are similar to the recorded results in other studies (21). When acetylcholine esterase is inactivated, it leads accumulation of acetylcholine throughout the nervous system and causes hyperstimulation of muscarinic and nicotinic receptors (22).

Accumulation of free radicals resulted from acetylcholine esterase activity inhibition leads to lipid peroxidation (23). Free radicals contribute in toxicity of pesticides which may persuade oxidative stress resulting in releasing of free radicals and modification in antioxidants or oxygen free radical scavenging enzyme system (24). In a study, Handekari *et al.* (19), reported that cells constantly

suffer from oxidative stress in spite of over activity of antioxidant defense mechanism indicated by increase in erythrocyte. Increase in antioxidant enzymes levels may be essential to purify increased concentration of lipid peroxidation yields that are oxidative stress products because of organophosphorus poisoning (23).

The significant increase in leucocytes observed after dipping in pesticides implies the activation of the defense mechanism and immunity. The same observation was reported by Hundekari (19) that the number of white blood cell count was increased significantly in all cases of OP poisoning. They also detected increased in total white blood cells with mild decrease in neutrophils count as compared to control values. In acute OP toxicity, overproduction of reactive O₂ species immediately results in major lipid peroxidation. The liability of erythrocytes and leucocytes especially lymphocytes to oxidative stress due to exposure to the pesticide is a function of overall equilibrium between degree of oxidative stress and antioxidant protection proficiency. Thus the antioxidant attack aptitude of exposed subjects may alter directly or indirectly by OP compounds and therefore affect their liability to oxidative stress of exposed subjects and consequently affect their vulnerability to oxidative stress (24).

There is significant decline in the mean values of hemoglobin concentration of animals after dipping compared to the mean values before dipping, possibly due to the decline in formation of hemoglobin or may be due to binding of organophosphorus insecticides on iron, followed by a lack of fusion of iron with hemoglobin leading to low MCV, i.e. decreased size of erythrocytes (microcytosis).

Conclusion

From the above results it was concluded that organophosphorus compounds cause a significant decline in AChE activity and the severity of poisoning aids in predicting OP poisoning and effects on blood parameters. These effects may be through oxidative damage caused by elevating lipid peroxide levels.

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Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

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التغير في نشاط الأستيل كولين وبعض معايير الدم في الأغنام المغطسة في مبيد دلتاميثرين

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الخلاصة

هدف الدراسة هو توضيح تأثير مركب الفسفور العضوي المستخدم في تغطيس الأغنام على أستيل كولين وبعض معايير الدم للأغنام من خلال تقييم التغيرات في مستويات أستيل كولين استريز في المصل. أجريت الدراسة على أربعين غنما غطست في حوض يحتوي على مبيد دلتاميثرين بتركيز 1:1000 لتر من الماء. تم جمع نماذج الدم الوريدي من كل حيوان قبل وبعد التغطيس لتقييم معايير الدم ومقايضة أستيل الكولين. أدلت النتائج الى وجود تثبيط معنوي في أستيل كولين استريز في جميع الحيوانات الخاضعة بعد الغطس. كما أظهرت القيم المتوسطة للمقاييس الدموية ارتفاعاً ملحوظاً في عدد الكريات البيض الكلية مع انخفاض معنوي في تركيز الهيموغلوبين ومؤشرات الخلية الحمراء في جميع الحيوانات بعد التغطيس في محلول المبيد عند مقارنتها مع القيم التي تم الحصول عليها من نفس الحيوانات قبل الغطس. كشفت النتائج التي توصلنا إليها أن التسمم مع دلتاميثرين يؤثر على معايير الدم من خلال تثبيط أستيل استريز.