ANALYSIS OF PESTICIDES RESIDUES IN DRINKING WATER AND HUMAN BLOOD IN MANSOURA CITY

By

Abdel-Aziz Ghanem, Somaia M. El-Azab, Raafat A. Mandour* and Mona S. EL-Hamady

*Department of Forensic Medicine and Clinical Toxicology , Faculty of Medicine, and Toxicology Lab, Emergency Hospital *Mansoura University, Egypt*

ABSTRACT

Pesticides are classified into four major groups; insecticides, herbicides, rodenticides and fungicides. Insecticides poisoning is one of the most common poisoning affecting our population. The aim of our study , is to investigate the relationship between cholinesterase inhibitor insecticides in human blood and insecticides residues in drinking water in Mansoura city. Sixty patients suffering from insecticides poisoning admitted to Emergency Hospital Mansoura University in March 2005, were included in this study. Blood samples were collected from these patients. The studied patients and their relatives were asked to obtain water samples of their drinking water from the localities where they live. Thirty drinking water samples were collected. Blood and water samples were extracted using Liquid-liquid extraction technique (LLE) and analyzed for pesticides using Gas Chromatography-Mass Selective Detector (GC-MSD). In addition, blood samples were analyzed for plasma pseudo-cholinesterase level (PChE) and red blood cells acetyl cholinesterase activity (AChE). The analysis of drinking water and blood samples revealed organophosphorous (Mevinphos) and organochlorine (Aldrin) insecticides. Moderate insecticides poisoning was more common among the studied groups (65.0%) compared to mild and severe poisoning. The peak incidence of poisoning was in the age group from 20-30 years (46.67%), followed by the age group from 10-20 years (43.33%). There was a female sex predominance among the poisoned patients (56.67%). There was a negative correlation between the severity of poisoning and each of PChE levels and AChE activity % being lowest (p<0.001) in severe poisoning compared to moderate and mild poisoning.

INTRODUCTION

Drinking water resources in Dakahlyia governorate are either surface from Nile River and the related irrigation canals or subsurface resources raised from some boreholes drilled in the Quaternary aquifer at Mit-ghamr, Aga and Sinbillawin districts. Abd el-Daiem and Ramsussen (1991) reported that at Mit-ghamr, the surface water level is almost +9 m where it is about +6 m for the surrounding groundwater level. Herein there is downward movement of the surface water to recharge the groundwater from the influent stream in this sector. On contrary, at Mansoura, the surface water level in the Nile branch is +1.8 m, while it is about +4 m in the nearby water wells, where the river behaves as a drain.

It is recognized that the environmental degradation products of pesticides may be a problem in drinking water, (WHO, 1997; Auersperger et al., 2005). Acute poisoning by pesticides is common; it usually occurs due to careless use, misuse or occupational exposure (Wang et al., 2006). According to EEC (1980) and WHO (1993), the presence of toxic pollutants in water especially insecticide chemicals can change the quality of surface water. In order to monitor the quality of surface water, samples must be analyzed periodically to assess the residues levels and distribution of chemical pollutants in the surface water.

Insecticides residues determination in water requires analysis at or below ppb level (µg/l), so the investigation of water pollution with such low concentration requires a concentration step prior to the analysis. Concentration step is often necessary to raise the concentration of the insecticide chemicals (found in the water at trace levels) above the sensitivity levels of the detection of the analytical tools. Liquid-liquid extraction technique (LLE) has been considered as an effective method for concentration of trace organic pollutants including insecticides from water samples (WHO, 1997).

The aim of the present work is to investigate the relationship between cholinesterase inhibitor insecticides in human blood and insecticides residues in drinking water at Mansoura city.

SUBJECTS AND METHODS

Sixty patients were admitted to the Poison Unit at Emergency Hospital, Mansoura University during March 2005. Patients were presented with signs and symptoms of cholinesterase inhibitor insecticides poisoning, with a variant degree of severity including sweating, diarrhoea, vomiting, excessive salivation, miosis, breathing problems, heart dysrrhythmias and extreme anxiety. These patients were enrolled in our study. A history was taken from the patients through answering a questionnaire, which included; age, sex, route and mode of exposure to the insecticides.

Blood samples were collected from all patients (n=60). The studied patients and their relatives were asked to obtain water samples of their drinking water from the localities where they live. Thirty drinking water samples were collected. The blood and water samples were analyzed for determination and identification of pesticides by GC-MSD. Moreover the blood samples were investigated for the levels of pseudo-cholinesterase enzyme and acetyl cholinesterase activity by a colorimetric method.

Extraction of water and blood samples, (Flanagan et al., 1995):

* pH of each sample (10 ml) was adjusted to 7 by adding solid sodium bicarbonate.
* Each sample was extracted with 5 ml of methyl tertiary-butyl ether for 5 minutes using a rotary mixer.
* Samples were left to stand for 5 minutes, then the upper layers were taken and re-extracted with a second 5 ml portion of methyl tertiary-butyl ether.
* The two extracts of each sample were filtered through a Wattmann filter paper No 4 into a clean tube and the extract of all samples were evaporated to dryness under compressed air.

Residues of the evaporated extracts were reconstituted by 100 µl chloroform. 1 µl of each extracted sample was injected into GC-MSD (Hewlett Packard 6890 series) of ECD (Electron Captured Detector) under the following conditions, (Maurer and Weber, 1992):

* Carrier gas (He)
* Capillary column; model No : HP19091Z-102, Hp-1 Methy Siloxane, length 25m, diameter 200 µm, film thickness 0.33µm.
* Flow rate 1.0 ml / min, velocity 39cm / sec, Mode: split less,
* Pressure 16.1 psi, Run Time 12.5 min
* Thermal Aux 2 (MSD; Mass Spectrum Detector), temp 280°C,

Max.temp. 325 °C, oven temp 200°C.

Determination of pseudo-cholinesterase enzyme levels and acetyl cholinesterase enzyme activity of blood samples:

Levels of pseudo-cholinesterase enzyme (PChE) and acetyl cholinesterase (AChE) activity were analyzed by spectrophotometer according to Ellman et al. (1961) and Crane et al. (1970), respectively.

Statistical Analysis :

The statistical analysis of data was done by using Excel and SPSS programs statistical package for social science version 10. The description of the data was done in the form of mean (+/-) SD for quantitative data and frequency & proportion for qualitative data. The analysis of the data was done to test statistical significant difference between groups. For quantitative data, one way ANOVA test was used to compare more than 2 groups , and for qualitative data Chi-square test was used. (N.B: p is significant if $\leq$ or = 0.05 at confidence interval 95%).

RESULTS

The analyzed drinking water and blood samples revealed the presence of organophosphorous (Mevinphos) and organochlorine (Aldrin) insecticides. The mass spectra of fragment ions are; 127, 192, 109 for Mevinphos (Fig. 1) and 66, 263, 79, 91, 101 m/z for Aldrin (Fig. 2).

Table (1) showed that the peak incidence of poisoning by cholinesterase inhibitor insecticides was in the age group from 20-30 years (46.67%), followed by the age group from 10-20 years (43.33%). (Fig. 3).

Table (2) showed that there was a female sex predominance among the poisoned patients (56.6%) (Fig. 4).

Table (3) showed that the moderate poisoning of cholinesterase inhibitor insecticides was more common among the studied group (65.0%) compared to mild and severe poisoning (Fig. 5).

Table (4) showed that there was a negative correlation between the severity of poisoning and each of PChE levels and AChE activity %. The cholinesterase enzymes showed highly significant decrease ($p<0.001$) in severe poisoning compared to moderate and mild poisoning.

DISCUSSION

The present work was carried out to investigate the relationship between cholinesterase inhibitor insecticides in human blood and insecticides residues in drinking water, in Mansoura city. The analyzed drinking water and blood samples by GC-MSD revealed the presence of organophosphorous (Mevinphos) and organochlorine (Aldrin) insecticides. This indicates no treatment for pesticides in the drinking water stations. However, the exposure to insecticides through a pollutant drinking water represent a rare source of exposure among other different sources including agriculture, industrial, vector control, and domestic purposes (Robert and Aaron, 2007). This explained the clinically variant degree of severity among the poisoned patients that was confirmed by cholinesterase enzymes assay.

As regard age, the study showed that (46.67%) were in the age group from 20 to less than 30 years (Table 1). This finding is in agreement with the result found by (Agarwal, 1993), who reported a peak incidence of poisoning by cholinesterase inhibitor insecticides in the 3rd decade of life. Moreover, 43.33% of patients were in age group from 10 to less than 20 years, this correlates with age ratio reported by (Marey, 1986). There was a female sex predominance among the poisoned patients (56.67%) (Table 2). This result is in agreement with the result of Bhattarai et al., (2006).

Compared to mild and severe poisoning, moderate poisoning of cholinesterase inhibitor insecticides was the commonest among the studied group (65.0%) (Table 3). There was a negative correlation between the severity of poisoning and each of PChE levels and AChE activity %, being lowest in severe poisoning compared to moderate and mild poisoning (Table 4) as proved by (Abdel-Magid and Salem, 1993; Jacobsen et al., 2004).

Presence of Aldrin in drinking water is attributed to contamination from industrial effluents and soil erosion during irrigation, where River sediments may contain higher amounts (up to 1 mg/Kg) (WHO, 1989). Existence of Aldrin in the investigated drinking water samples represents a serious environmental problem for human. According to WHO (1993) and EMOHR (1995), Aldrin is very dangerous organic pollutant in surface water where the target organs being the central nervous system and the liver. It has been regulated as a possible human carcinogen primarily on the basis of animal studies, however, the epidemiologic evidence is inconsistent (Purdue el al., 2007). On the other hand, this compound induced immunosuppressive and hormonal disruption activities in animals and humans (Thomas, 1995; Colborn et al., 1996).

CONCLUSION

Drinking surface water in the area of study was polluted with organophosphorous and organocholrine insecticides compounds. The blood of patients complaining of cholinesterase inhibitor insecticides poisoning showed the same insecticides which were detected in the drinking water. Presence of these pollutants in surface drinking water of the studied area has serious effects on human health and environment.

RECOMMENDATIONS

The following recommendations would be considered by local authority in Dakahlyia governorate:

1. Multiple-stage filters containing granular activated carbon as an adsorbent material (with replaceable cartridges) could be used mainly in drinking water stations and in houses.

2. Chemical analyses could be carried out periodically for the surface and groundwater to ensure the water suitability for drinking purposes (water must be free from pesticide residues or within the permissible limit of WHO).

3. Complete knowledge of organophos-

phorous and organochlorine insecticides residues and their transformation products in surface water is one of the most important factors for solving the environmental problem related to these chemicals.

4. Authorities must prohibit the use of organocholrine and restrict the use of organophosphorous insecticides.

Table (1): Patients (n=60) of acute cholinesterase inhibitor insecticides poisoning as regard the age of peak incidence

Age groups	Poisoned patients	
	(n)	%
10- 20	26	43.33
20^+ - 30	28	46.67
30^+ - 40	3	5.0
40^+ -50	3	5.0
Total	60	100.0

Table (2): Patients (n=60) of acute cholinesterase inhibitor insecticides poisoning as regard the sex predominance.

Sex	Males		Females		Total	
	(n)	%	(n)	%	(n)	%
Poisoned patients	26	43.33%	34	56.67%	60	100.0

Table (3): Patients (n=60) of acute cholinesterase inhibitor poisoning as regard the degree of poisoning.

Degree of poisoning	Poisoned patients	
	(n)	%
Mild	15	25.0
Moderate	39	65.0
Severe	6	10.0
Total	60	100.0

Table (4): Comparison between the degree of poisoning and mean values (U / l) and activity percentage of cholinesterase enzymes.

Cholinesterase Enzymes	Mild poisoning >1000	Moderate poisoning 1000 -500	Severe poisoning < 500		P Value
(PChE)Mean±SD	1301.43±352.51	578.55±176.62	205.82 ± 81.74	F = 13.87	P<0.001***
(AChE) Activity%	46.37%	20.56%	7.27%	$X^2 = 24.5$	P<0.001***

P***: highly significant

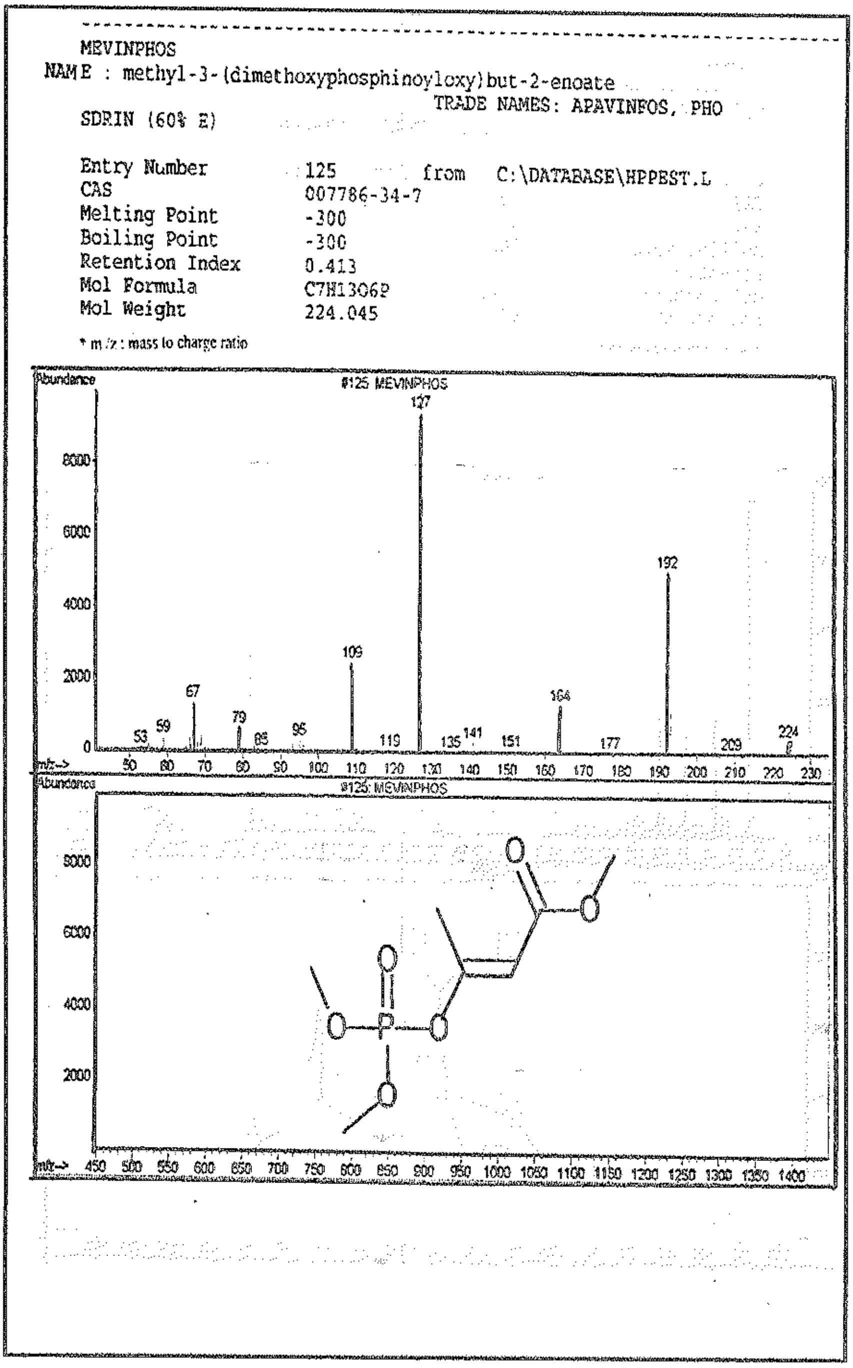

Fig. (1): Mass spectrum and structure of Mevinphos.

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ALDRIN
    (1R,4S,4aS,5S,8R,8aR)-1,2,3,4,10,10- hexachloro-1,4,4a,
5,8,8a-hexahydro- 1,4,5,8-dimethanonaphthalene
                    TRADE NAMES: OCTALENE

Entry Number        49        from   C:\DATABASE\HPPESI.L
CAS                 000309-00-2
Melting Point       -300
Boiling Point       -300
Retention Index     1
Mol Formula         C12H8Cl6
Mol Weight          361.875
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* m/z : mass to charge ratio

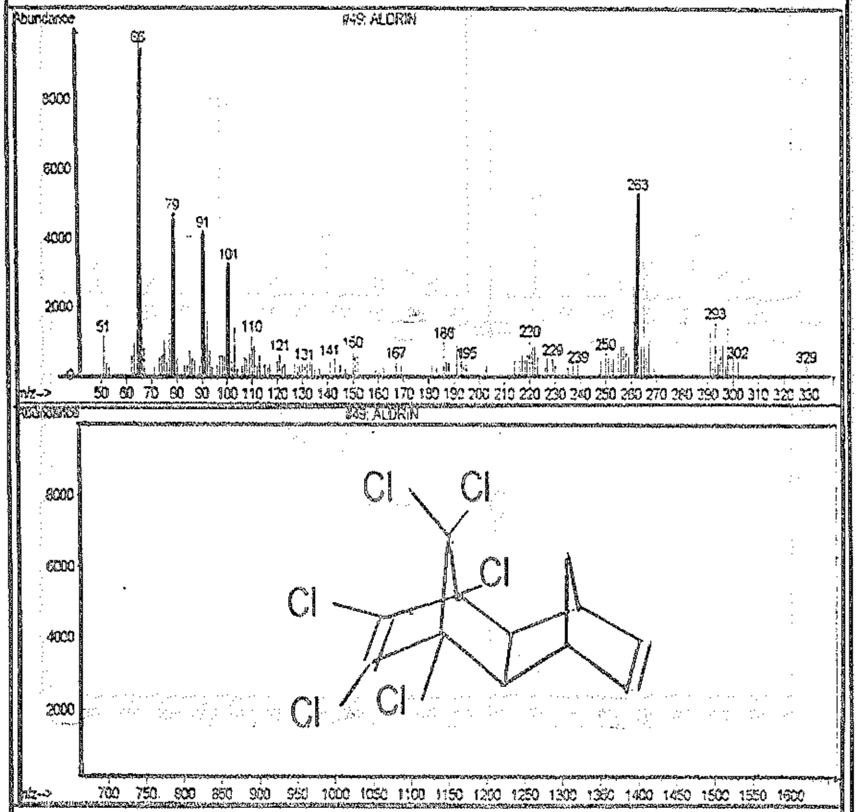

Fig. (2): Mass spectrum and structure of Aldrin.

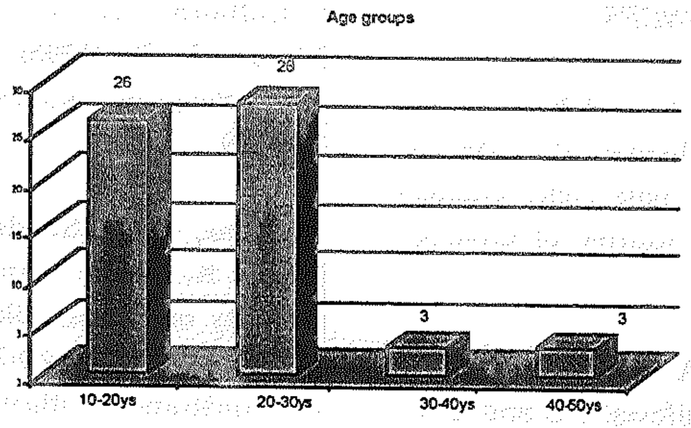

Fig. (3): Acute cholinesterase inhibitor insecticides poisoning as regard the age of peak incidence.

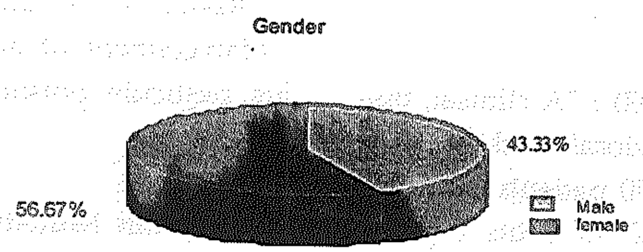

Fig. (4): Acute cholinesterase inhibitor insecticides poisoning as regard the sex predominance.

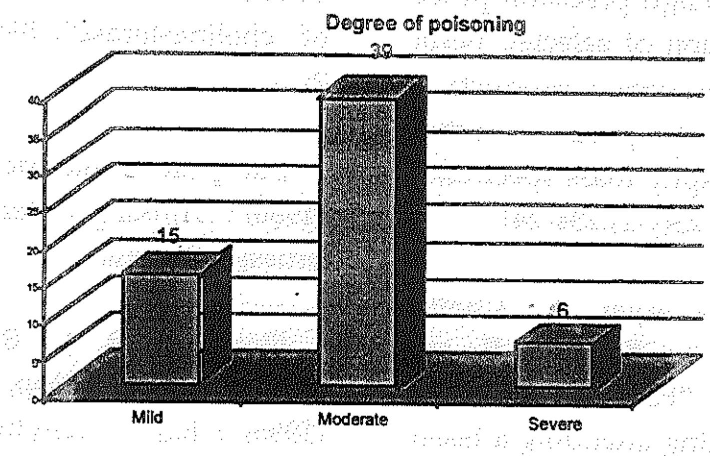

Fig. (5): Acute cholinesterase inhibitor insecticides poisoning as regard the degree of poisoning.

REFERENCES

Abd el-Daiem, A. A. and Ramsussen, K. R. (1991) : "A mathematical model for the north part of the Nile Delta Quaternary aquifer, Egypt". Journal of Environ. Sci., 3:339-363.

Abdel-Magid, L. A. and Salem, E. M. (1993) : "Clinical manifestations and management of patients with acute poisoning by anticholinesterase insecticides (Anti-ChEI)". J. Pest. Control & Environ. Sci., 5 (2): 1-21.

Agarwal, S. B. (1993) : "A clinical, biochemical, neurobehavioral and socipsychological study of 190 patients admitted to hospital as a result of acute organophosphorous poisoning". Environmental Research, 62: 63-70.

Auersperger, P. ; Lah, K. ; Kus, J. and Marsel, J. (2005) : "High precision procedure for determination of selected herbicides and their degradation products in drinking water by solid-phase extraction and gas chromatography-mass spectrometry". J. Chromatogr., 23(1-2):234-241.

Bhattarai, N.; Rauniyar, A.; Chaudhary, D.; Jaiswal, S.; Banthia, P. and Rana, B. B. (2006) : "Patterns of organophosphorous poisoning attending a teaching hospital". J. Nepal. Med. Assoc., 45 (162): 228-232.

Colborn, T.; Dumanoski, D. and Myers, J. P. (1996) : Our Stolen Future. Pengum Books, USA, NY, P.P. 189-209.

Crane, C. R.; Sanders, D. C. and Abbott, J. K. (1970) : Studies on the Storage Stability of Human Blood Cholinesterase. I. Report No. AM 70-74 Federal Aviation Administration, Office of Aviation Medicine, Civil Aero Medical Institute, Oklahoma City.

Robert, D. M. and Aaron, C. R. (2007) : "Management of acute organophosphorus pesticide poisoning". BMJ., 334 : 629-634.

Egyptian Ministry of Health Regulations (EMOHR) (1995) : Report No. 108 (in Arabic).

Ellman, G. E.; Fest, J. S. H. and Gross, T. L. (1961) : "Colorimetric determination of cholinesterase". Biochem. Pharmacol., 21: 7.

European Economic Community (EEC) (1980) : Drinking water directive (80/779), Brussles, Belgium.

Flanagan, R. J.; Braithwaite, R. A.; Brown, S. S.; Widdop, B. and Wolff, F. A. (1995) : Basic Analytical Toxicology. 1st ed., World Health Organization, Geneva, P.P. 188 - 189.

Jacobsen, H.; Østergaard, G.; Lam, H. R.; Poulsen, M. E.; Frandsen, H.; Ladefoged, O. and Meyer, O. (2004) : "Repeated dose 28-day oral toxicity study in Wistar rats with a mixture of five pesticides often found as residues in food: alphacypermethrin, bromopropylate, carbendazim, chlorpyrifos and mancozeb". Food Chem. Toxicol., 42(8):1269-1277.

Marey, H. M. (1986) : Toxicological study on cases of organophosphorous insecticides admitted to the emergency department of Alex. Main University Hospital. MSc emergency medicine, Faculty of Medicine; Alex. University.

Maurer, H. H. and Weber, A. (1992) : Mass Spectral and GC Data of Drugs, Poisons, Pesticides, Pollutants and Their Metabolites. VCH, Weinheim, NewYork, Basle, 2nd ed., part 1.

Olson, K. R. (1994) : Poisoning and Drug Overdose. Prentice-Hall International, Inc., USA, P.P. 118-240.

Purdue, M. P.; Hoppin, J. A.; Blair, A.; Dosemeci, M. Alav and anja, M. C. (2007) : "Occupational exposure to organochlorine insecticides and cancer incidence in the Agricultural Health Study". Int. J. Cancer, 120(3):642-649.

Thomas, P. T. (1995) : "Pesticides - induced immunotoxicity- are the great lakes residents at risk". Environ. Health Prospect., 103(9): 55-61.

Wang, D. X.; Zhu, X. L.; Zhu, G. Z. and Du, X. Q. (2006) : "The etiology and rescue modalities of 1692 cases of acute chemical poisoning". Zhonghua Nei Ke Za Zhi, 45 (8):631-634.

World Health Organization (WHO). (1989) : Aldrin and dieldrin, (environmental health criteria, No 91), Geneva, Switzerland.

World Health Organization (WHO). (1993) : Guidelines for drinking water quality. 2nd ed., Vol.1, Geneva, Switzerland.

World Health Organization (WHO). (1997) : Guidelines for drinking water quality. 2nd ed., Vol.2, Health criteria and other supporting information. Geneva, Switzerland.

تحليل متبقيات المبيدات الحشرية فى مياه الشرب ودم الإنسان بمدينة المنصورة

المشتركون فى البحث

أ. د. عبدالعزيز أبوالفتوح غانم   د.سميه محمد العزب
د. رأفت عبدالغنى أبومندور   د. منى صلاح الدين الحمادى

قسم الطب الشرعى والسموم الإكلينيكية*، كلية الطب - جامعة المنصورة

معمل السموم بمستشفى الطوارىء* - جامعة المنصورة

يعتبر تحليل ملوثات المبيدات الحشرية فى مياه الشرب من الأمور الهامة لحماية البيئة والحفاظ على صحة الإنسان من أخطار هذه المبيدات.

يهدف هذا البحث إلى دراسة العلاقة بين متبقيات المبيدات الحشرية فى مياه الشرب ودم الإنسان بمدينة المنصورة.

أجرى هذا البحث على 60 من المرضى يعانون من التسمم بالمبيدات الحشرية بدرجات متفاوتة وقد أدخلوا وحدة السموم بمستشفى الطوارىء - جامعة المنصورة خلال شهر مارس ٢٠٠٥.

تم الحصول على عينات دم من جميع المرضى (60 عينة) كما تم تجميع ٣٠ عينة من مياه الشرب بواسطة هؤلاء المرضى ومرافقيهم من مناطق سكنهم.

تم تحليل عينات الدم ومياه الشرب وذلك على جهاز التحليل الكروماتوجرافى - مطياف الكتلة للوقوف على مدى تلوثها بمتبقيات المبيدات الحشرية.

كما تم تحليل عينات الدم لقياس مستوى تركيز الكولين استريز.

وقد أظهرت النتائج وجود المبيد الحشرى الفسفورى العضوى (ميفنفوس) والمبيد الحشرى الكلورينى العضوى (الألدرين) فى عينات الدم ومياه الشرب.

وكانت حالات التسمم على أساس مستوى تركيز الكولين استريز تتفاوت بين حالات تسمم شديد وبسيط ومتوسط، والذى كان يمثل النسبة الأكبر بين الحالات (65.0%).

وقد يعزى ذلك التفاوت لتعدد مصادر الإصابة بالتسمم بالمبيدات الحشرية بجانب مياه الشرب.

كما أظهرت النتائج وجود علاقة إرتباط عكسية بين مستوى تركيز الكولين استريز فى الدم ودرجة شدة الإصابة بالتسمم حيث كان مستوى تركيز الكولين استريز أقل بين حالات التسمم الشديد (205.82±81.74) بالمقارنة مع حالات التسمم المتوسط والبسيط على التوالى (578.55±176.62) و (1301.43±352.51).

وقد أوصت الدراسة بالآتى :

١- إستخدام فلاتر المياه المتعددة المراحل والتى تحتوى على الكربون النشط المحبب بالمنازل ومحطات مياه الشرب وعمل تحاليل دورية لمياه الشرب.

٢- زيادة التوعية بخطورة المبيدات الحشرية المثبطة لعمل الكولين استريز لما لها من تأثير خطير على الإنسان.

٣- منع إستخدام المبيدات الحشرية الكلورينية والحد من إستخدام المبيدات الحشرية الفسفورية.