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Serum Cholinesterase Level Reduction in Pesticide Factory Workers

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In this study the sampling from 63 subjects was carried out for two sessions in days 1 and 90. Cholinesterase determination was performed with butyrylthiocholine substrate and enzymatic test was done using kinetic techniques. The mean serum cholinesterase level in phase 1 was 9569 ± 2496 IU L⁻¹ and in second phase the activity of cholinesterase was 7970 ± 2067 IU L⁻¹. This drop in cholinesterase level statistically was meaningful (paired t-test, mean = 1599, 95% CI = 1140-2058, p<0.001). ALT increase in second phase compared to first phase statistically was meaningful (Paired t-test, mean = -7.9, 95% CI = -10.9 -4.9, p<0.001). In this study eight subjects (12.7%) had more than 35% reduction in cholinesterase activity. In regard to reduction in cholinesterase activity of 17 workers which include 27% of working personals in a three months period it seems logical to set a program to have a routine check on the cholinesterase activity in working personal engaging in such occupations.

Key words: Pesticide, organophosphate, cholinesterase, Iran, serum

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INTRODUCTION

Cholinesterase (ChE) is one of the important enzymes required for the proper function of nervous system. Human contain two type of ChE, red blood cell ChE (EC 3.1.1.7) or true-cholinesterase and plasma ChE (EC 3.1.1.8) or false cholinesterase (Thetkathuek *et al.*, 2005).

Some of the chemical constituents of pesticide such as organophosphate and carbamate either interfere with the activity or inhibit of this enzyme. Breathing, eating, absorption through the skin and eyes are the routes that ChE inhibitor can poison human (Oakeshott *et al.*, 2005). It is estimated 3 million people are poisoned by organophosphate toxins annually and about 300000 of such poisoned subjects either are dying or having serious damage (Eyer, 2003). The ChE level in a subject after poisoning return to its normal level after 50 days, if the person after the accident does not come into contact with the toxins again (Mason, 2000).

The determination of serum ChE level is a good indicative of acute status of organophosphate toxin contacts. It is advisable that every working personal in the pesticide factory should have their ChE level to be determined before starting the job and this level should be considered as a base level for every individual. This research designed for detection of changes SChE level in workers that were exposed to organophosphate pesticides in 3 months period.

MATERIALS AND METHODS

This study is a cohort study. This study was carried out during 2005 on the pesticide factory workers in Golestan province (North of Iran, Southeast of Caspian Sea). A questionnaire was filled for every employee and 3 mL of blood from the participant of this research was taken. Their blood samples were transferred to the laboratory, the serums were separated using the centrifuge and the separated serum was stored at -70°C. During two phase of experiment which lasted for 3 months, 63 working personal of the factory went under this investigation.

Albumin measurement was carried out by Bromocresol green (BCG) technique. AST and ALT measurement was carried out by IFCC (international federation of clinical chemistry) using Pars azmoon company kit (Iran) by photometer (clinic II). Serum cholinesterase (SChE) test was carried out by Pars azmoon kit with butyrylthiocholine substrate. The confidence interval for all the tests was considered to be 95%.

RESULTS

All factory workers were men. The minimum age of the study population was 21 year and the maximum age was 53 (mean 32±8.03 years). The working personal were divided in five groups according to variation of ChE level. The first group includes the subjects with either no reduction or increase with their ChE level. ChE level in second group, third group, fourth group and fifth group had a reduction of less than 15, 15-25, 26-35% and more than 35 %, respectively (Table 1). On the basic of place of work, in the factory working personal they were divided in 5 groups. Eighteen people (28.6%) in service technical and administrative section, 16 people (25.4%) in store and packaging, 12 people (19%) in powder and liquid fertilizer section, 12 people (19%) in herbicide and pesticides section and 5 people (7.9%) in primary material production section. There was not any meaningful correlation between reduction of SChE level and place of work (Table 2).

The experience of factory personal was between 0-7 years (Mean 2.59±1.95). According to record of service the employee were divided in 4 groups. Thirteen people (20.7%) less than one year, 19 people (30.6%) 1-3 years, 19 people (30.6%) 3-5 years and 12 people (19.1%) more than five years. There was not significant meaning between experience and SChE reduction.

The minimum and maximum level of SChE level in phase one were 4960 and 15420 IU L⁻¹, respectively. The mean level SChE level in phase one was 9569±2496 IU L⁻¹. the minimum and maximum of SChE level in phase two were 3700 and 13020 IU L⁻¹, respectively. The mean level of SChE in phase two was 7970±2067 IU L⁻¹ (paired t-test, mean = 1599. 95% CI = 1140-2058, p<0.001).

The mean of AST activity in phase one was 22.8±8.8 IU L⁻¹ and in phase two 24.7±10.1 IU L⁻¹. The mean AST in phase one and two statistically did not have any meaningful differences. The mean level of ALT in phases one and two were 16.6±8.3 and 24.5±13.2 IU L⁻¹, respectively (paired t-test, mean = -7.9, 95% CI = -10.9-4.9, p<0.001). The serum Albumin concentration in phase one 4.51±0.040 g dL⁻¹ and in phase two 4.01±0.78 g dL⁻¹ (paired t-test, mean = -0.50, 95% CI = 0.27- 0.73, p<0.001).

Table 1: The prevalence of serum cholinesterase variation in factory workers

Group	N	(%)
Without reduction Or increase	14	22.2
Less than 15%		
<15%	16	25.4
15-25%	16	25.4
26-35%	9	14.3
More than 35%		
>35	8	12.7
Total	63	100.0

Table 2: The prevalence of cholinesterase level reduction according to the place of work in the factory

Place of work	Without reduction or increase	<15% reduction	15-25% reduction	25-35% reduction	>35% reduction	Total
Service, technical	6.0	5.0	5.0	1.0	1.0	18
Percentage in the group	33.3	27.8	27.8	5.6	5.6	100
Storing and package	5.0	4.0	3.0	3.0	1.0	16
Percentage in the group	31.3	25.0	18.8	18.8	6.3	100
Powder and liquid fertilizer	0.0	2.0	1.0	5.0	4.0	12
Percentage in the group	0.0	16.7	8.3	41.7	33.3	100
Herbicide and insecticide	1.0	5.0	6.0	0.0	0.0	12
Percentage in the group	8.3	41.7	50.0	0.0	0.0	100
Production	2.0	0.0	1.0	0.0	2.0	5
Percentage in the group	40.0	0.0	20.0	0.0	40.0	100
Total	14.0	16.0	16.0	9.0	8.0	63
	22.0	25.4	25.4	14.3	12.7	100

Mean of Body Mass Index (BMI) of the working personals was $25.56 \pm 5.15 \text{ kg m}^{-2}$. There was not any meaningful correlation between BMI and SChE reduction. Altogether 8 subjects (12.7%) had a reduction of more than 35% in their SChE level.

DISCUSSION

The determination of ChE activity has a high application in the diagnosis of liver damaged by pesticide (Osawa *et al.*, 2005). One of the groups which are exposed to the risk of chronic poisoning by the pesticide is the personal working in the pesticide producing factories.

Organophosphate and pesticide reduce the ChE activity. In this study the sampling was done twice in period of three month study. Some times the ChE activity is in normal rang, but it still shows a meaningful reduction, therefore in this study every sample participates was his own control and the determination of ChE activity in phase one was considered the base level for a particular worker. In a study in Iran (Mashad) by Vahdati *et al.* (2004) was concluded that 70-80% of Iranian people are carrier of a mutation on the ChE gene allele (Vahdati *et al.*, 2004). Also in another study Housaini *et al.* (1997) have shown that there is significant difference on ChE activity between the normal Iranian and Irish population (7.82 ± 0.14 vs. 5.22 ± 0.09 , $p < 0.01$) These finding indicate the importance of ChE level measurement in the society and the determination of basic level for every worker, prior to work in places exposed with the toxins.

In a study on a Mexican farmers it was found that there was a significant differences between ChE activity before starting and at the end of the job season (paired $t = 2.7$, $p = 0.01$) (Lopez-Carillo and Lopez-Cervantes, 1993).

In a study of Innes and colleagues in South Africa which was carried out on 44 of fruit corps spraying, it was found that 7 out of 44 people had sever reduction of plasma ChE. The BMI of these 7 people was lower than

others (Innes *et al.*, 1990). But in present study there was not a meaningful correlation between SChE level and BMI.

In a study done in Malaysia by Husin *et al.* (1999) on the farmers workings in rice farm land, there was a meaningful reduction in the SChE level. A study in California on 542 people who were in charge of applying the pesticide the SChE level of 26 subjects (4.8% of the total sample population) was 50% of lower base level of ChE activity (Ames *et al.*, 1989).

In an experimental study, dogs were fed by fertilizer and 50% reduction of ChE level was observed (Yeary, 1984). This finding has a correlation with the results of this present study, which showed the highest rate serum ChE reduction level was among worker, engaged in the production of primary products, powder production and liquid fertilizer.

AST and ALT serum activity increase and Albumin concentration reduction is an indicative of damage to the liver cells by pesticide contact. In an experimental study, it was shown that Methidathion cause a meaningful increase in AST, ALP (Alkaline phosphatase), GGT (gamma- glutamyltransferase) and LDH (lactate dehydrogenase) in cases, but ALT activity is lowered compared to the controls (Altuntas *et al.*, 2002). The results of this experiment were totally different from the present study. In present study although the AST activity was increased during a three months working period, but this increase statistically did not have any meaning, but ALT increase was obviously clear. In a study by Kalender *et al.* (2005) on Wistar rats Albumin, AST and ALT had a visible increase after a four weeks period by given Diazinon (organophosphate insecticide). These findings are similar to our results from the present study.

In regard to the reduction of SChE level by 25% in 17 working personal (27%) of the pesticide factory of this study, routine ChE measurement seems an obligatory process in such occupations.

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