

BIOLOGICAL MONITORING BY SERUM CHOLINE ESTERASE IN DIFFERENT OCCUPATIONAL GROUPS EXPOSED TO PESTICIDES

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Abstract

Background: Pesticide are used extensively in agriculture to enhance food production by eradicating unwanted insects. Pesticide can cause harm to human, animals, or the environment. The aim of this study to evaluate the biochemical changes in human of different occupational groups exposed to pesticide. **Material and methods:** The cross sectional comparative study was conducted in the department of biochemistry Chattogram Medical College, from January 2012 to December 2012. Total 100 subjects who fulfilled the enrollment criteria were included in this study. Among them 60, who were exposed to pesticides were included in group A or case or test subject and 40 who were not exposed to pesticides were included in group B or control. The occupational groups exposed to pesticide were randomly selected on the basis of their active involvement at least 5 years in preparation, storage and spraying of the pesticides. **Results :** The present study showed that serum Cholinesterase(ChE) was significantly lower in cases than that of control with $p = <0.001$. In this study observed that 45% cases decreased ChE level. Regarding serum SGPT, it is significantly increase in cases than that of control (45.05 ± 16.08 vs 31.85 ± 4.25 U/L, $p = <0.001$). It is also observed that nearly 45% cases increased SGPT and SGOT level in cases(Group A). **Conclusion:** It was found that there is a significant decrease in serum cholinesterase in insecticide spraying workers while serum SGPT and SGOT were increased in exposed group. It is

thus concluded from the findings of the study that clinicians and epidemiologists should advice occupationally exposure group to periodic screening test for serum ChE, SGPT, SGOT to detect the effect of pesticides before adverse health effect to occur.

Key words

Pesticides; Choline esterase; SGPT; SGOT.

Introduction

A pesticide is any substance or mixture of substances intended for preventing, destroying, repelling, controlling the pest and insects in the environment so as to protect human health and agricultural output. Pesticides can cause harm to humans, animals, or the environment because they are designed to kill or otherwise adversely affect living organism^{1,2}. Pesticides are used extensively in agriculture to enhance food production by eradicating unwanted insects. The wide spread use of pesticide in agriculture has caused environmental pollution and potential health hazards. Pesticides produce adverse biological effects through the active ingredients and associated impurities. Pesticide exposure represents a major potential health hazards for sprayers. Major chemical groups of pesticides are Organophosphate (OP) carbamate, organochlorines, nitro and chlorophenols and pyridyl derivatives^{2,3}. A hazards can be defined as a source of danger. The hazards in using a pesticide is related to the likelihood of exposure to harmful amounts of pesticides. A pesticide can be highly toxic as a concentrate, but pose to little hazards to the user if a pesticide may have relatively low toxicity but present a hazards because it is used in the concentrated form which may readily absorbed or inhaled⁴. Now organophosphorus insecticides are extensively used for the control of all types of insects pests⁵. Because of long residual effects, agricultural use of organochlorines have been restricted in many countries including Bangladesh. However, at present these organophosphorus compounds are the major cause of pesticide poisoning. Several studies confirmed the presence of breakdown product of OP in the urine sample. This indicates that most people are routinely exposed to

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these chemicals (U.S Centers for Disease Control (CDC) and Academic scientists)^{6,7}.

It has been observed that maximum exposure occurs to workers who mix, load or apply pesticides (Known as pesticides handlers) due to spills, splashes, defective, missing or inadequate protective equipments, direct spray, or drift contact with pesticide residues on the crop or soil^{8,9,10,11}. Wind is the single most important factor determining dermal exposure during spraying. Other factors are equipment used, duration of exposure, types of activity and individual protection^{12,13}.

The biological effects produced by certain pesticides can be enzyme induction or enzyme inhibition. The effect of pesticide may be detected by ensuring biochemical changes even before advance clinical health effects can occur. The effects of chronic exposure do not appear immediately after first exposure and may take years to produce symptoms. Exposures to pesticides cause a reduction of Choline esterase (ChE) activity, which can be used for their risk assessment¹⁴. Cholinesterase is an enzyme present in plasma and synthesized by the liver. The primary biological role of ChE is the rapid hydrolysis of Acetylcholine at the cholinergic synapses in the central and peripheral nervous system and at neuromuscular junctions. The inhibition of ChE by certain xenobiotic chemical (eg. organophosphorus pesticide) within synaptic clefts results in the dysfunction of nerve transmission by preventing the inactivation ChE; this leads to excessive stimulation of CNS/PNS. Altered liver enzyme activities have been reported among pesticide workers exposed to OP alone or in combination with organochlorine or other pesticides^{15,16}.

Biological monitoring by determination of blood ChE activity has important role in assessing the exposure of workers to OP pesticides. Blood cholinesterase testing is done to evaluate potential human exposure to chemicals that act as ChE inhibitors, most often OP and carbamate pesticide are inhibitors of cholinesterase^{17,18}. Prolong exposure to pesticide affect multiple organs including liver which can be detected by serum enzymes and other biochemical parameter. Pesticides exposure causes leakage of cytosolic enzymes from hepatocytes and other organs^{18,19}.

Continuous exposure to mixtures of pesticides including OPs in farmers engaged to intensive agriculture leads to signs of cytotoxicity resulting in

early biochemical changes, such as in serum enzymes and other standard serum parameters. Another important findings from Bangladesh and also in Brazil is that, specific crops and geographic locations experience more overuse than others. OP pesticides have gained popularity worldwide in preference to organochlorines, which are persistent and more damaging to the environment^{19,20}.

A higher population of pesticide poisoning and death occur in developing country like Bangladesh where there are inadequate occupational safety standards, protective cloths and washing facilities, insufficient enforcement, poor labeling of pesticides, illiteracy and insufficient knowledge of pesticide hazards²¹.

There has been no study so far on the biochemical aspects of environmental health with specific reference to prolong exposure to pesticides from developing countries. Information on how pesticides affect health is quite limited in many developing countries, with many surveys relying solely on farmers and sprayers self- assessments of their health status^{21,22}. Therefore we have planned to study the biochemical effects of the pesticide pollution on human health.

Materials and methods

The cross sectional comparative study was conducted in the Department of Biochemistry, Chattogram Medical College from January 2012 to December 2012.

Total 100 subjects who fulfilled the enrollment criteria were included in this study. Among them 60, who were exposed to pesticides were included in group A or case or test subject and 40 who were not exposed to pesticides were included in group B or control. Again group A were subdivided into Group A1 (Farmers) and Group A2 (Sprayer).

Inclusion criteria

The occupational groups exposed to different pesticides were randomly selected on the basis of their active involvement at least 5 years in preparation, storage and spraying of the pesticides and control group were age matched and ranged from 20 – 55 years.

Exclusion criteria

The farmers on the basis of giving history of suffering from diabetes mellitus, chronic renal failure, toxic or viral hepatitis, chronic liver diseases,

malignant metastasis, any other chronic diseases not related to pesticide exposure, taking alcohol and female were excluded from the study to avoid any interference with biochemical parameters measured.

Data were collected by interview of the subjects, clinical examination and laboratory investigations using the research instrument.

Blood samples (5ml) were collected into clean and dry deionized test tubes and transported to laboratory within icebox to prevent haemolysis. The blood sample were kept for 30 minutes for separation of serum and then centrifuged at 3000 rpm for 5 minutes and biochemical analysis was done in deionized test tube and serum choline esterase, SGPT, SGOT was measured as per photometric colorimetric method by using specific kits [Photometer 5010].

Serum cholinesterase level-4659–14443 U/L, SGPT level- 0- 42U/L, SGOT level – 0 - 42/L.

Data was analyzed by computer based software SPSS V 15. Data were expressed as mean ± SD. Confident level was fixed at 95% level and “p” value of 0.05 or less was considered significant. Student’s ‘t’ test for quantitative or continuous variables, Chi-square test for categorical variables were done where applicable. The summarized data were presented in the form of tables and charts.

Results

The findings obtained from data analysis were presented below:-

Table I : Distribution of Serum cholinesterase among cases (Group A) and Control (Group B) (With t- test significance)

Study Group	n	Mean	±SD	Range	Significance
Group A1	40	5225.90	3032.40	1293-12207	t= 0.373 p= 0.710 Not significant
Group A2	20	5544.75	3284.99	1292-11203	
Group A	60	5332.18	3094.58	1292-12207	t = 4.787 p= 0.000
Group B	40	7861.85	2187.88	4942-12920	Very highly Significant

Table I showed that Serum cholinesterase was significantly decreased in cases than that of control (5332.18 vs 7861) with p = < 0.001.

Table II : Distribution of serum cholinesterase in Group A and Group B as number and percentage (With χ^2 significance)

Serum Cholinesterase Status	Study Groups						Total	
	Group A 1 (Farmer)		Group A 2 (Sprayer)		Group B (Control)		n	%
Normal	22	55.0	11	55.0	40	100.0	73	73.0
Decreased	18	45.0	09	45.0	00	0.0	27	27.0

χ^2 value = 24.658. p = 0.000. Very highly significant.

Table II showed that serum cholinesterase was decreased 45% in cases than that of control with p= < 0.001.

Table III : Distribution of SGPT in cases (Group A) and Control (Group B) (With t- test significance)

Study Group	n	Mean	±SD	Range	Significance
Group A1	40	46.47	16.08	21- 92	t= 0.273
Group A2	20	42.20	14.18	24- 78	p= 0.810
Group A	60	44.33	15.13	21- 92	t = 3.687
Group B	40	31.85	4.25	24- 39	p = 0.005 Very highly Significant

Table III showed that in cases mean SGPT is more increase than that of control (44.33 vs 31.85) with p= 0.005).

Table IV : Distribution of SGPT in cases (group A) and control (Group B) as number and percentage with χ^2 significance.

Serum SGPT Status	Study Groups						Total	
	Group A 1 (Farmer)		Group A 2 (Sprayer)		Group B (Control)		n	%
Normal	22	55.0	12	60.0	40	100.074	74.0	
Increased	18	45.0	08	40.0	00	0.0	26	

χ^2 value = 23.597. p = 0.000. Very highly significant.

Table IV showed that nearly 45% cases increased SGPT than that of control with p value < 0.001.

Table V : Distribution of SGOT in cases (Group A) and control (Group B) with t-test significance

Study Group	n	Mean	±SD	Range	Significance	
Group A1	40	40.98	15.08	21-76	t=0.473	
Serum SGOT (U/L)	Group A2	20	40.00	12.18	24-76	p=0.510
					Not significant	
	Group A	60	40.46	15.13	21-76	t=5.587
	GROUP B	40	26.63	6.05	17-39	p=0.001
					Very highly Significant	

Table V showed that in cases mean SGOT is more increase than that of control (40.46 vs 26.63, p= 0.001).

Table VI : Distribution of SGOT in cases and control as number and percentage (With χ^2 significance)

Serum SGOT Status	Study Groups							
	Group A 1 (Farmer)		Group A 2 (Sprayer)		Group B (Control)		Total	
	n	%	n	%	n	%	n	%
Normal	22	55.0	12	60.0	40	100.0	74	74.0
Increased	18	45.0	08	40.0	00	0.0	26	26.0

χ^2 value = 23.597. p = 0.000. Very highly significant.

Table VI showed that in cases (Group –A) 45 % subjects increase SGOT with p value < 0.001.

Discussion

The purpose of the study was to determine serum cholinesterase and SGPT, SGOT for assessment of adverse effects in hepatocellular functions in different occupational groups exposed to pesticides for routine clinical and epidemiological purposes.

In this study the ChE activity have been assayed in different occupational groups that are routinely exposed to pesticides. The biochemical study also extended to evaluate the toxicology of pesticides in human and in this regard liver enzymes (SGPT, SGOT) have been assayed.

In this study all the test subjects were male. Female are excluded from the study as ChE activity is greatly changed during pregnancy. The mean age of the test subjects were 39 ± 7 years.

Several studies on volunteers reported that repeated long term exposures of organophosphorus pesticide and carbamate decrease the blood cholinesterase activity without clinical manifestation^{22,23}.

The present study showed that serum cholinesterase was significantly lower in case than that of control (5332.18 ± 3094.58 vs 7861.85 ± 2187.88 U/L, $p < 0.001$). Similar results were noted among the cotton growers and tobacco farmers of many countries due to chronic pesticide exposure^{23,24}.

In this study observed that 45% cases decreased serum ChE level, but in control group not decreased, which is consistent with many other studies^{24,25}. The reduction of ChE activity in pesticide exposed workers in present study demand official regulations and interventions to protect workers dealing with pesticides from the adverse health effects of these chemicals. Several studies on volunteers reported that repeated long term exposure of organophosphorus and carbamate pesticides decrease blood ChE activities without clinical manifestation²⁵. Blood Cholinesterase activity has been used for several years to estimate the risk associated with pesticide induced toxicity in occupationally exposed workers.

Regarding serum SGPT, it is significantly increase in cases than that of control (45.05 ± 16.08 vs 31.85 ± 4.25 U/L, $p < 0.001$). It is also observed that nearly 45% cases increased SGPT level in study population. A similar observation was found in other studies^{25,26}.

Regarding serum SGOT level, significantly increase in cases than control group (40.65 ± 14.03 vs 26.63 ± 6.06 U/L, $p = 0.001$). About 43% cases increase SGOT level in this study. These result was consistent with study done on agriculture and farm workers²⁶. The increase in the level of SGPT and SGOT is a good indicator of hepatic toxicity.

One Way ANOVA analysis shows that incase of farmers and sprayers significantly decrease serum ChE level than that of control, $p = < 0.001$. The SGPT and SGOT increased in farmers and sprayers than that of control (46 and 40 vs 26). A similar observation was observed in other studies²⁷.

Pearson's correlation coefficient shows a negative relation between serum SGPT, SGOT and cholinesterase, $p = < 0,001$. A similar observation was found in others studies²⁸.

Pesticides are widespread used in agricultural sectors in Bangladesh and most of the agricultural workers, and sprayers (Environmental disinfectant workers) in city corporation area are at risk due to careless handling of pesticides. High degree of abnormal liver function in agriculture workers may indicate toxic effects of pesticide and presence of pesticide residue in blood. Greater the degree of pesticide exposure, greater will be the level of liver enzymes.

However, further a large study is required in this regard because the small sample size and selected subjects from some area of Bangladesh may not be representative of whole Bangladesh.

Conclusion

The present study was undertaken to assess the impact of pesticides on occupationally exposure group to find out serum cholinesterase and its correlation with SGPT, SGOT for assessment of adverse effect in hepatocellular functions. It was found that there is a significant decrease in serum cholinesterase in insecticide spraying workers while serum SGPT and SGOT were increased in exposed group. It is thus concluded from the findings of the study that clinicians and epidemiologists should advice occupationally exposure group to periodic screening test for serum ChE, SGPT, SGOT to detect the effect of pesticides before adverse health effect to occur. This study will pave the way for early diagnosis and monitoring of pesticide induced adverse effects among occupational exposure by biochemical markers.

Contribution of Authors

MKH - Conception, acquisition of data, data analysis, drafting and final approval.

SANMK - Data analysis, Critical revision of content and final approval.

MH - Design, interpretation of data, critical revision of content and final approval.

Disclosure

All the authors declared no conflict of interest.

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