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Enzymatic Liver Changes among Workers Exposed to Vinylchloride

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Abstract

Background Polyvinyl chloride (PVC) is used in production and manufacturing of many essential tools for example plastic pipes, fabric, cables, decorative products etc.). Its production is impossible without the use of vinyl chloride monomer (VCM), which can cause liver damage in long-term.

Objective To assess the effects of mild to moderate long term exposure to VCM on liver and to assess the importance of liver enzyme measurements as screening tools.

Methods In this study, measurement of serum levels of liver enzymes of 64 exposed workers and 61control workers was carried out starting from the first of October 2010 till the end of January 2011. All of the studied cases were worked in a poly vinyl chloride (PVC) production unit in three polyvinyl chloride factories and considered as target population for detection of any possible industrial vinyl chloride associated liver enzymes changes. The controls were randomly selected from office personnel of the same factories. Biochemical paramedics and a questionnaire method were used for analysis and in both groups.

Results Both groups have a similar age structure. Statistical difference was noted between the alanine aminotransferase (ALT) and gamma-glutamyl transferase (GGT) mean values for both the exposed and non-exposed groups. The mean values for alpha-2-globulin and gamma-globulin in both exposed and non exposed groups of serum electrophoresis were statistically significant. The relative risk for the exposed workers was higher than that one for all other variables. It was the highest and most significant for gamma-globulin abnormal values associated among the exposed group followed by the relative risk of alpha-2- globulin.

Conclusion Liver function tests with serum protein electrophoresis are useful to detect hepatic damage among workers exposed to polyvinylchloride.

Key words Liver Enzymes, Workers, Protein Electrophoresis, Vinyl Chloride

Introduction

Vinyl chloride (VC) is a colorless organic gas with a sweet odor, and is used to make polyvinyl chloride (PVC) plastic and vinyl products [(1,2)]. It is used in the manufacture of numerous products in building, construction, the automotive industry, electrical wire insulation, cables, piping, industrial and household equipment, medical supplies, rubber, paper, and glass industries [(1,2)].

VC is a known human carcinogen (cancer-causing agent) [(3)]. VC is also a known genotoxicant, causing chemical alterations of DNA in tissues that may lead to cancer

following exposure of humans and experimental animals [3]. The primary target organ for VC exposure is the liver [4]. The association between angiosarcoma of the liver and vinyl chloride exposure is well documented for occupational exposures [4]. Noncancer liver pathologies have also been associated with VC exposure, including liver necrosis and cysts [4]. Several studies in experimental animal models have demonstrated that early life exposure to VC can increase susceptibility to cancer later in life [5]. VC is a synthetic chemical used as a chemical intermediate in the polymerization of PVC [6]. At room temperature and pressure VC is poorly soluble in water. Structurally, VC is a haloalkene and is related to vinylidene chloride and trichloroethylene. Human and animal data indicate that VC is rapidly and efficiently absorbed via the inhalation and oral routes, is rapidly converted to water-soluble metabolites, and is rapidly excreted. At low concentrations, VC metabolites are excreted primarily in urine, while at high exposure concentrations; unchanged VC is also eliminated in exhaled air. Overall, the data indicate that neither VC nor its metabolites are likely to accumulate in the body.

Absorption of VC in humans after inhalation exposure is rapid. A study conducted in five young adult male volunteers inhaling VC at concentrations of 7.5 to 60 mg/m^3 showed that 42% was retained, maximum retention was reached within 15 minutes, and the percent retention was independent of inspired VC concentration [7].

VC is produced on a substantial scale - approximately 31.1 million tons were produced in 2000[7]. An important subject in health preservation of workers exposed to VCM is the early detection of their effects. Unfortunately minor liver damages can be detected through routine screening tests such as aminotransferase measurement and needs more specific tests such as the measurement of others liver enzymes level [8]. It is used in the manufacture of personal protective equipment such as in the gloves material for hand protection used by the forensic scientists during crime scene investigation as it is resistant to alkalies, oil and limited concentration of nitric and chromic acids [9].

Several studies have been conducted on the detection of early effects of VCM on workers with contradictory results in many factories where the PVC workers are exposed to below the threshold levels of VCM [10]. Regarding the increase in the number of such workers in our country and no study was conducted about the problem, we designed this study.

Methods

A cross sectional study was carried out to determine the prevalence of enzymatic liver changes among exposed and unexposed workers, 64 exposed workers were compared to 61 control_workers during September 2010 through February 2011. All cases were working in a PVC production unit in three PVC factories of the national chemical and plastic industries which was established in 1983 and situated in Zafarania\Baghdad and considered as target population for detection of any possible industrial VC associated liver enzymes changes. The controls were randomly selected from office personnel of the same factories. This factory is specialized in the production of PVC granules (total product 85tons/day) which are used in decorative products, cables, houses and fabric industry.

Data collection

After explaining the objectives of the study to the workers and taking their verbal consent, the data were collected from the workers by using specially constructed questionnaire.

Demographic data gathered in the questionnaire include age, sex, marital status, weight, height, work experience, alcohol consumption, tobacco smoking, past medical history, drug history, performing heavy exercises, work history including any changes

of the job and second job, history of surgery and history of blood transfusion .

A thorough clinical examination, signs and symptoms , with special attention to the signs that may be related or associated with liver disease such as jaundice, clubbing of fingers, palmer erythema, spider naevi, ascites, hepatomegally and splenomegally. The blood samples were drawn from the workers through a venepuncture 1.0 ml of the blood was added to 0.2 ml of 0.11 molar sodium citrate in a test tube to be used for the determination of Prothrombin time (PT). The remaining part of the blood sample was allowed to coagulate, centrifuged and the serum separated was divided and stored into three labeled test tube at 20^{o}C for other measurement , the first one for the determination of total serum bilirubin (using the method of Malloy with the normal range being 0.2-1.0 mg /100ml), alanine transaminase (ALT), aspartate transaminase (AST) and alkaline phosphatase (ALP); the second one for gamma glutamyl transferase (GGT); and the third one for total serum protein and protein electrophoresis. The serum was deep freeze until used for these parameters ,usually within less than 48 hours.

Methods used for Biochemical investigations in the study:

a) Total serum bilirubin ;
b) ALT: The spectrophotometric methods of Reitman and Frankel (normal range of 2-20 I.U/L).
c) AST: The spectophtometric methods of Reitman and Frankel (normal range of 2-15 I.U/L).
d) GGT: GGT reagent cartridge kit (normal range 7-64 I.U/L) [11].
e) ALP: Spectophtometric assay was used (normal range being 3-14 K.A.U/100L) [12].
f) PT: This was done by the Quick one stage method (normal range of 10-13 second) [13].
g) Total serum protein Electrophoresis: The Biuret method for the determination of total protein in serum (normal range of 62-77 g /L) [14].

The serum electrophoresis was carried out according to normal values: albumin = 35-50 g/L, α_1-gloubulin = 1-4g/L, α_2-gloubulin = 4-8g/L, β-globulin =5-10 g/L and γ-globulin =60 - 13 g/L. All these biochemical investigations were performed in the factories of National Center of Occupational Health and Safety in Baghdad.

Data analysis*:* was done by using:

a. Descriptive statistic: tables (frequency and percentage)
b. The relative risks (RR) with their 95% confidence intervals (CI) were estimated [15].
c. Inferential statistic: t-test was used to test the statistical differences between group means (Minitab version 13)

Result

Sixty four workers occupationally exposed to VC were studied and compared with 61 non-exposed workers. Both groups have a similar age structure (Table 1) with mean of 36.79 ±8.60 years for the exposed and a mean of 37.52± 8.85 years for the non exposed workers. No statistical difference could be detected between the two age means (p>0.05).

The majority 31 (48.88%) of the exposed workers and 29 (45.31%) of the non exposed workers fall in the age group 30-39 years. All workers (exposed and unexposed were males), and all of them were Iraqis. The mean duration of employment for the exposed workers was 5.53±3.51years. Twenty nine (45.31%) have a duration of employment of 1-5 years and quite a large number 23 (35.94%) have a duration of employment 6- 10 years (Table 2). Table 3 shows positive clinical findings relevant to the liver disease in VC exposed and non exposed workers .Hepatomegally was detected in 7. 81% of the exposed and in 3.28% of the non exposed workers .Exposure to VC carries more than twice the risk for developing hepatomegaly (RR= 2.38, 95% CI=0.48 – 11.8).

Splenomegaly was found in 1.56% of the exposed while none of the non exposed

workers had such a finding. Clubbing of the fingers was detected in 4.69% of the exposed and in 1.64% of the non exposed which carries a relative risk of 2.86 and a 95% CI of 0.31-26.58. None of exposed and non exposed workers was jaundiced at the time of examination. Spider naevi, palmer erythema, ascites and other signs of liver disease were not detected in any of the studied groups (Table 4).

Table 1. Age distribution of exposed and non exposed workers to vinyl chloride

Age groups (years)	Exposed workers (64)		Non-Exposed workers (61)		mean±Sd	T test	Level of significance
	No.	%	No.	%			
20-29	12	18.75	11	18.03	26.27±1	0.3	p>0.5
30-39	31	48.44	29	45.31	34.09±2.45	0.16	p>0.5
40-49	13	20.31	13	21.31	43.18±3.18	1.23	p>0.5
50-59	8	21.5	8	13.11	53.50±1.77	0.66	p>0.5

Table 2. Duration of employment for workers exposed to vinyl chloride

Duration of employment	Number of workers	Percent of total (n=64)
≤1 years	4	6.25
1-5 years	29	45.31
6-10 years	23	35.94
≥10	8	12.5

Table 3. Clinical findings relevant to liver disease in vinyl chloride exposed and none exposed workers.

Clinical findings	exposed workers (64)		Non-exposed workers (61)		Relative risk	95% CI for Relative risk
	No.	%	No.	%		
Hepatomegally	5	7.81	2	3.28	2.38	0.84-11.83
Splenomegally	1	1.56	0	0	-	-
clubbing fingers	3	4.69	1	1.64	2.86	0.31-26.58

Table 4. Past medical history and symptoms relevant to liver disease in vinyl chloride exposed and non-exposed workers

Medical history and symptoms	exposed workers (64)		Non-exposed workers (61)		Relative risk	95% CI for Relative risk
	No.	%	No.	%		
Jaundice	2	3.13	1	1.64	1.91	0.18-20.49
Upper abdominal discomfort	17	28.56	9	14.75	1.80	0.87-3.74
Loss of appetite	8	12.50	5	8.20	1.53	0.53-4.44
Nausea	7	10.94	5	8.20	1.33	0.45-3.92
Loss of weight	3	4.69	1	1.64	2.86	0.31-26.58
Hepatitis	3	4.96	2	3.28	1.43	0.25-8.33

Table 5 shows a statistically significant difference in the ALT and GGT mean values between exposed and non exposed groups (p< 0.05). Other test i.e. total serum bilirubin, AST, ALP, and PT showed no statistically significant difference in both study groups(p> 0.05), although such values were all higher in the exposed than the non-exposed groups.

Table 5. Liver function tests in vinyl chloride exposed and none exposed workers

Liver Function Tests	Exposed workers n=(64)	Non-exposed workers N=61	T test	P value
Total serum bilirubin	0.44±0.17	0.43±0.11	0.39	P>0.05
ALT	10.06±7.39	7.10±4.89	2.65	P<0.005
AST	10.24±4.69	9.70±4.00	0.69	P>0.05
GGT	34.62±35.96	23.23±15.55	2.32	0.05>P>0.01
ALP	9.74±5.27	8.73±3.74	1.23	P>0.05
****PT**	13.03±0.26	13±0	0.88	P>0.05

* This result applies to 58 workers only

Table 6 shows the mean values for different components of serum protein electrophoresis. There was statistically significant difference between α_2-globulin mean values for both the exposed and non exposed groups (p< 0.05 and p<0.005 respectively). The mean concentration of the total protein, albumin, α_1-globulin, and β-globulins, where not statistically significantly different in both study groups (p> 0.05).

Table 6. Serum protein electrophoresis and total protein in vinyl chloride exposed and none exposed workers

Serum protein electrophoresis and total protein	Exposed workers n=(64)	Non-exposed workers N=61	T test	p- value
Albumin	3.99±0.38	4.02±0.34	-0.47	P>0.05
α_1-globulin	0.31±0.14	0.29±0.05	1.07	P>0.05
α_2-globulin	0.71±0.14	0.67±0.08	1.97	0.05>P>0.025
β -globulin	0.91±0.23	0.89±0.14	0.59	P>0.05
γ-globulin	1.57±0.77	1.13±0.27	2.54	P<0.05
Total protein	7.39±0.50	7.25±0.45	1.65	P>0.05

Table 7 illustrate the relative risk of exposed group ranged between 1.27 for AST and 2.38 for GGT with other values for total serum bilirubin, ALT and ALP falling in between. All 95% confidence intervals built around such relative risks had lower limits of less than one.

Table 8 shows that the relative risk for the exposure was higher than one for all variables being the highest and most significant (RR=3.81, 95% CI=1.12- 13.07) for γ-globulin abnormal values associated with exposure followed by that of α_2-globulin (RR=3.34, 95% CI=0.72-15.33).

Table 7. Rates for abnormal components of serum protein electrophoresis in vinyl chloride exposed and none exposed workers

Serum protein electrophoresis	exposed workers (64)		Non-exposed workers (61)		Relative risk	95% CI for Relative risk
	No.	%	No.	%		
Albumin	7	10.94	3	4.92	2.22	0.60-8.25
α_1-globulin	2	3.31	1	1.64	1.91	0.18-20.49
α_2-globulin	7	10.94	2	3.28	3.34	0.72-15-33
β -globulin	3	4.69	2	3.28	1.43	0.25-8.25
γ-globulin	12	18.75	3	4.92	3.81	1.21-13.07

Table 8. Liver function tests abnormality (rate percent in vinyl chloride exposed and none exposed workers)

Liver Function Test	exposed workers (64)		Non-exposed workers (61)		Relative risk	95% CI for Relative risk
	No.	%	No.	%		
Total serum bilirubin	2	3.13	1	1.64	1.97	0.18-21.12
ALT	9	14.06	4	6.56	2.14	0.70-6.55
AST	4	6.25	3	4.92	1.27	0.30-5.47
GGT	5	7.81	2	3.28	2.38	0.48-11.8
ALP	7	10.94	3	4.92	2.22	0.60-8.25
PT	1	1.75	0	0.00	0.00	0.00

Discussion

Clinical symptoms associated with vinyl chloride exposure were mainly that of digestive manifestations including anorexia, nausea, abdominal distention, epigasteric pain, pain in the right and left hypochodrium, and loss of weight [16]. In our study low values of digestive manifestations may be explained on the basis of low exposure levels, and that digestive manifestations associated with high exposure levels are sometimes accompanied by some neurological manifestations [17]. Such neurological manifestations were totally absent in our study. Clinical features related to liver disease that could mainly be associated with VC-exposure include hepatomegally, splenomegally, hematemesis and melena, jaundice, spider naevi, palmer erythema and ascites [18]. The low rate of hepatomegally in this study may also due to effect of transfer from one to another section in the studied factories because workers in these factories move around among different places at different intervals during the year depending on factory needs and priorities, hence they might be exposed to different (usually lower) concentration of VC than actually suggested by their working shift time. In our study only 1.56 % of exposed workers had splenomegally and none had the history of haematemesis or melena as well as spider navei, palmer erythema, jaundice and ascities were not found on clinical examination. So our result showed lower rates than those shown by other studies and such high rates in other studies may be related to more advanced stage of the disease that might have not existed in our result [19].

In the present study only 3.13 of the exposed workers had elevated total serum bilirubin also our study has demonstrated that 14.1% of the exposed workers have elevated ALT. Elevated AST were found in 6.26% of the studied cases.

Elevated ALP values were recorded in 10.9% of the exposed study workers.

The abnormalities of total serum bilirubin, ALT, AST and ALP of this study are generally lower as compared to the other studies [20]. Such discrepancy could be explained by shorter period of exposure, discontinuity of exposure and lower level of VC. In addition, the surveyed workers in other studies were usually selected on the basis of clinical manifestations beside the effect of social factors such as alcohol intake where 21.88% of our exposed workers had history of alcohol intake while others studies had high rate of alcohol intake [21]. Regarding PT and GGT, no comparable studies could be found to compare our result with. In this study there is a significant difference between the mean values of ALT and GGT for the VC in exposed and non exposed workers while no significant difference was found concerning values of total serum bilirubin, AST, ALP and PT. The significant difference of ALT may be explained on the basis that VC or it is metabolites. Also significant elevation of ALT and GGT may indicate that the effect of vinyl chloride or it is metabolites are mainly on the liver cells because mechanism of toxicity and carcinogenity of VC is hypertrophy and hyperplasia of hepatocytes and sinusoidal dilatation and destruction, hepatocyte destruction, portal tract fibrosis and binding of coloroethylene oxide (VC metabolite) to DNA and RNA [22].

For PT significant difference was not found between exposed and non exposed workers and this may be attributed to the fact that the liver changes are not so advanced to disturb the synthetic function of the liver to the extent to cause decreased clotting factors synthesis .In our study values for components of serum protein electrophoresis were done for both of studied groups. It is obvious clear to note that all of our results are lower that obtained by another studies. [22] Such different between our study and another studies may explained by more advanced liver changes which is produced by longer and higher level of exposure to VC, also such higher result in another studies could be explained by the selection of those workers [23].

In our study significant difference was found between VC exposed and non exposed workers for the means values of α_2-globulin and γ-globulin, such findings could be explained by the presence of liver injury by VC or it is metabolites [23]. Elevated γ-globulin level could be noticed whenever there is prolonged and marked immune response [24] considered VC disease an immune complex disease.

Conclusions

Our results indicate that although the laboratory results were all within normal range but liver involvement in PCV processing workers is still possible and should be given full attention in the medical surveillance of the workers. Our results showed that laboratory tests were of limited values in the identification of VC associated liver disease, but it is wise to run the usual battery of tests annually for the sake of early detection of changes that could accompany other findings detected by other methods of investigation Such as ultrasonagraphy, computerized tomography, serum bile acid levels and Indocyanin clearance test [25].

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