BIOCHEMICAL AND RADIOISOTOPIC EVALUATION OF LIVER FUNCTION IN WORKERS EXPOSED TO VINYL CHLORIDE

M. GLUSZCZ and Z. MYŚLAK

Institute of Occupational Medicine, Clinic of Occupational Diseases and Acute Intoxications, Łódź, Poland

The authors examined liver functions in 60 persons with Raynaud--Syndrome exposed to vinyl chloride (VC). Some patients were exposed to VC till the moment of hospitalization, the others had 1–4 years break in exposure to VC. In order to assess the liver function, the following biochemical tests were applied: activity of alanine and aspartate amino-transpherase, gamma-glutamyl--transpeptidase, lactic dehydrogenase, alkaline phosphatase in serum. Morover the following tests were used: BSP test, determination of prothrombin time, bilirubin in blood, level of free and esterified form of cholesterol and of lipoproteins.

Simultaneously a hepatography was made using ^{131}I and scintigraphy using bengal rouge or colloid ^{99m}Tc. It was found that biochemical tests revealed the liver lesion in a small number of persons exposed to VC. In the same patients, it was found, that a proportionally greater group had positive scintigraphic analysis tests i.e. elongated time of partial marker disappearance in blood and an average liver enlargement. The authors are of the opinion that the scintigraphic method may be more useful in detection of liver lesion caused by exposure to VC than the biochemical tests.

LONG TERM MORTALITY STUDY OF VINYL CHLORIDE AND POLYVINYL CHLORIDE WORKERS IN A JAPANESE PLANT

Y. MASUDA

Health Administration Center, Keio University, Tokyo, Japan

In order to investigate the health problem resulting from the past vinyl chloride (vc) exposure, a long term mortality study of vc workers and controls was carried out.

All number of workers emloyed in vc and pvc plant amounted to more than 304 since the beginning of vc production. A total of 304 workers exposed to vc for more than 1 year after 1949 (vc workers) and 277 men employed at other facilities (controls) have been traced and deaths from 1949 through 1975 were ascertained. Of the vc workers, 218 men experienced operation of polymerization or reactor cleaning and the others were assigned to vc synthesis, shipping or maintenance. The concentration of vc in the working environment was cotroled under 500 ppm for about 5 years before 1960 and since then under 250 ppm until 1974.

The expected number of deaths were calculated on 5-years age/time cause specific mortality for Japanese males and 5-years age/time classified number of workers with person years from 1956 to 1975. Distribution of cause of deaths was tabulated using a copy of death certificates and information from doctors in charge or families.

The observed number of deaths from all causes almost agreed with what was expected in vc workers and controls. The ratio of observed to expected ranged 0.44–5.00 for vc workers and 0.45–4.29 for controls. 38% of excess death from cancer was oberved in vc workers without statistical significance. Deaths from liver cirrhosis were 5 times as many as expected, which did not suggest the origin related to the vc exposure comparing with the ratio among controls. Anyhow prevention of liver cirrhosis is noted as an important problem among all these workers.

THE EFFECT OF MATERNALLY INHALED VINYL CHLORIDE ON EMBRYONAL, FETAL AND POSTNATAL DEVELOPMENT IN RATS

E. MIRKOVA, A. MICHAILOVA, L. HINKOVA and M. NASKO

Institute of Hygiene and Occupational Health, Medical Academy, Sofia, Bulgaria

The effects of vinyl chloride on embryogenesis and postnatal development of three generations of progeny in rats was studied. Vinyl chloride was applied daily to the pregnant rats from Day 1 through 20 of gestation in the inhalation chamber at a concentration of 6.15 mg/m³/day. Half of the animals were sacrificed on Day 21 of gestation. Fetal survival, number of resorptions, live fetal weights plus crownrump lengths and gross abnormalities by external examination were determined at necropsy. The fetuses were processed for examination of visceral neural and skeletal abnormalities. For the hygiene standardization the sensitive methods which allow an evaluation of biochemical deviations in the fetuses and functional changes in the offsprings were employed. The activity of dehydrogenases – LDH, MDH, SucDH, G_6PDH, i-CDH in the liver and the brain, the activity of γ-GTP and the concentration of soluble proteins in the liver were determined. For the evaluation of possible deviations in the nervous system of the offspirings the test »open field« was established.

The results shoved that vinyl chloride caused an increased fetal mortality and frequency of haemorrhages and a decreased fetal weight. Vinyl chloride was teratogenic at the concentration used. Teratogenic effects consisted of external (encephalocele), internal soft tissue abnormalities (hydrocephalus, brain haemorrhages) and skeletal malformations (doubles ossification centres in the sternum). Vinyl chloride produced also biochemical and functional disturbances in the homeostasis of the fetal organism and the offspring. A decreased activity of the dehydrogenases – G_6PDH, SucDH, i-CDH, MDH, LDH and an increased activity of γ-GTP was established in the fetal liver. Vinyl chloride caused an increased G_6PDH and i-CDH activity in the fetal brain. The index of fertility and gestation in the progeny was decreased. The excretory, detoxifying and enzymic functions of the liver, as well as the nervous system were injured in the three generations of the offspring.

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THE DYNAMICS OF VINYL CHLORIDE INDUCED ACROOSTEOLYSIS

Z. MYŚLAK and M. GŁUSZCZ

Institute of Occupational Medicine, Clinic of Occupational Diseases and Acute Intoxications, Łódź, Poland

The authors examined 306 workers exposed to vinyl chloride (VC) in a plant producing polyvinyl chloride with the suspension method. Among them there were 50 workers suffering from micro-vasels insufficiency, most of them with complete Raynaud-Syndrome. In 4 cases the Raynaud-Syndrome was accompanied with sclerodermia-like skin changes. In 4 cases acroosteolysis of hand bones was observed and in one person acroosteolysis of both fingers and toes. The time of exposure to VC before the diagnosis of acroosteolysis ranged from 3 to 5 years. The workers suffering from VC-induced disease were removed from their work in exposure to VC and they were periodically observed. It was noticed then that the bone changes were regressing. After a year a decrease of osteolysis was observed and after obout 2.5 years the X-ray picture of the hands and feet was normal, without any acroosteolysis. These observations show that in VC-induced osteolysis no real lesion of bone structure occurs, and that the acroosteolysis is reversible.

HISTOLOGICAL AND ELECTRON MICROSCOPE STUDIES UPON THE LONG--TERM TOXICITY OF VINYL CHLORIDE IN RATS, WITH SPECIAL REFERENCE TO LIVER CHANGES

J. STETKIEWICZ, J. KOŁAKOWSKI, I. ZDRAJKOWSKA and S. SZENDZIKOWSKI

Institute of Occupational Medicine, Łódź, Poland

Light-microscope studies were performed on internal organs and tissues including skin, skeletal muscles, bones, blood vessels and hemopoetic tissue of rats sacrificed after 1, 3, 6 and 10 months of exposure to vinyl chloride vapours in concentrations 50, 500 and 20,000 ppm. No evident pathological alterations were encountered in animals after exposure to vinyl chloride of 1 and 3 months durations. Histological changes were found in the liver of rats exposed to vinyl chloride at 500 and 20,000 ppm over the period of 6 months and in all three concentrations employed after the exposure of 10 month duration. The following changes were recorded:

1. Variable multifocal alterations in hepatocyte size ranging from decrease to marked cell hypertrophy, which becomes manifest and often insular after a longer time of exposure to both higher concentrations.
2. Parallel increase in nuclear polymorphism and in the number of binuclear hepatocytes.
3. Alterations in distribution and intensity of staining of the cytoplasmic basophile material.
4. An increased number of reticuloendothelial cells in the hepatic stroma.
5. Small focal cellular infiltrates composed mainly of lymphocyte – like cells, histiocytes and hemoblastic cells with occasional presence of bizzare megacariocytes.

Electron-microscope studies were limited to the livers. Following ultrastructural alterations were recorded: cytoplasmic hepatocyte changes – variations of mitochondrial electron density and apparent swollen mitochondria with elongated cristae, increase in number of microbodies and variations of their size mostly accompanying mitochondrial changes, alterations in endoplasmatic reticulum distribution, distention of rough reticulum canals and variable smooth reticulum hypertrophy, areas of focal cytoplasmic degradation; nuclear alterations – nuclear shrinkage and indentations with distention of perinuclear zone, increase in nucleolar size and number; Kupffer cells changes, represented by an increased phagosome numbers and by the presence of lipide droplets.

The histological and ultrastructural alterations observed are consistent with the criteria of liver lesions due to the other hepatotoxic substances described in the literature. They do not suggest any specific features of liver impairment under the long-term influence of vinyl chloride in the experimental conditions employed in this study.

EXPERIMENTAL STUDIES ON THE CHRONIC TOXIC EFFECTS OF VINYL CHLORIDE IN RATS

J. A. SOKAL, B. BARAŃSKI, J. MAJKA, R. ROLECKI, S. MATYCH, G. OLCZAK, J. PALUS, J. STETKIEWICZ, I. STETKIEWICZ and K. WRÓBLEWSKA

Institute of Occupational Medicine, Łódź, Poland

In the experiment, male rats were exposed to vinyl chloride at the concentrations of 50, 500 and 20,000 ppm, 5 hours a day, 5 days a week, during a total period of 10 months. After 1, 3, 6 and 10 months of the exposure, blood and urine analysis as well as histopathological examination of internal organs were performed in exposed and in control rats. Blood analysis included hematology and the following biochemical determinations: total protein, urea, phosphates, citrates, calcium, alkaline phosphatase, alanine and aspartate aminotransferases, γ-glutamyltranspeptydase and lactate dehydrogenase. In addition to the routine urine analysis acid mucopolysaccharides and hydroxyproline were estimated. After 10 months of exposure all rats were sacrificed, organ weights measured and the X-ray examination of the skeleton of some rats was performed.

As a result of chronic exposure to vinyl chloride the following toxic effects were observed in rats:

1. depression in body weight increase
2. increase in the relative weights of liver, kidneys, heart, and spleen
3. biochemical changes in blood (mainly increased activity of plasma lactate dehydrogenase and alkaline phosphatase)
4. slight hematological changes
5. morphological changes of the liver

Even at the concentration of 50 ppm some toxic effects were observed including morphological changes in the liver of some rats.

The results obtained showed that the chronic exposure to 50 ppm of vinyl chloride causes systemic toxic effects in rats.

HISTOLOGICAL AND ELECTRON MICROSCOPE STUDIES UPON THE LONG--TERM TOXICITY OF VINYL CHLORIDE IN RATS, WITH SPECIAL REFERENCE TO LIVER CHANGES

J. STETKIEWICZ, J. KOLAKOWSKI, I. ZDRAJKOWSKA and S. SZENDZIKOWSKI

Institute of Occupational Medicine, Łódź, Poland

Light-microscope studies were performed on internal organs and tissues including skin, skeletal muscles, bones, blood vessels and hemopoetic tissue of rats sacrificed after 1, 3, 6 and 10 months of exposure to vinyl chloride vapours in concentrations 50, 500 and 20,000 ppm. No evident pathological alterations were encountered in animals after exposure to vinyl chloride of 1 and 3 months durations. Histological changes were found in the liver of rats exposed to vinyl chloride at 500 and 20,000 ppm over the period of 6 months and in all three concentrations employed after the exposure of 10 month duration. The following changes were recorded:

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The histological and ultrastructural alterations observed are consistent with the criteria of liver lesions due to the other hepatotoxic substances described in the literature. They do not suggest any specific features of liver impairment under the long-term influence of vinyl chloride in the experimental conditions employed in this study.

METABOLISM OF VINYL CHLORIDE MONOMER IN THE LIVER OF RATS

J. M. WIŚNIEWSKA-KNYPL, S. TARKOWSKI and W. DRAMIŃSKI
Institute of Occupational Medicine, Łódź, Poland

Vinyl chloride monomer (VCM) undergoes biotransformation in the liver via oxidation by microsomal enzymes and/or conjugation with non-protein thiols catalysed by sulfotransferases (cf. Hefner et al., 1975; Green and Hathway, 1977).

In this experiment the following aspects of VCM metabolism have been assessed: (1) conjugation – on the basis of a decrease of -SH groups level in the liver and excretion of thiodiglycollic acid (TDGA) in the urine; (2) microsomes--mediated oxidation – on the basis of efficiency of the reaction of conjugation in phenobarbital and cobaltous chloride-pretreated rats.

Male Wistar rats were exposed by inhalation in dynamic chambers to VCM at concentrations of 50, 500 and 20,000 ppm 5 hours daily, for different periods of time.

It has been found within an hour after the end of exposure a marked decrease (by 19 and 54%) of -SH group content in the liver in rats exposed to 500 and 20,000 ppm of VCM, respectively. The level of the -SH group within 5–7 hours progressively recovered, reaching significantly higher values after 19 hours, than in the concurrent control.

Excretion of TDGA in the urine was dependent on the concentration of VCM but not on the duration of exposure.

Induction of synthesis of cytochrome P-450 in the liver of rats prior to the exposure to VCM resulted in much more pronounced decrease of -SH groups level in the liver immediately after exposure and increase of excretion of TDGA with urine. None of the decrease of the -SH group's content and deeply decreased level of TDGA were found in analysed materials from rats pretreated with the inhibitor of synthesis of cytochrome P-450.

It may be concluded that VCM first undergoes oxidation by cytochrome P-450 and only metabolites are conjugated.

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