

ABSORPTION, DISTRIBUTION AND ELIMINATION OF STYRENE IN MAN AND EXPERIMENTAL ANIMALS

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ABSTRACT

The authors studied the absorption, distribution and elimination of styrene both in man and in experimental animals by measuring its intake and concentration in blood and tissues. Volunteers inhaled the air containing a known concentration of styrene, the exhaled air was collected and styrene concentration was measured. Rats were exposed to styrene vapour in an exposure chamber at a controlled air flow rate of 2 l/min. The blood and tissues of rats were homogenized, styrene was extracted with N-hexane and analyzed by gas chromatography.

In man the rate of styrene intake by inhalation amounted to approximately 70%. In rats the constant level was reached about 180 minutes after exposure to styrene. After a single four-hour exposure of rats to styrene the concentration of styrene was highest in adipose tissue and lowest in muscles. The elimination of styrene from rats' tissues was expressed by an equation of the first order with a similar decline among the tissues except for the adipose tissue.

The latest development of the petrochemical industry has brought with it a health problem for synthetic resin workers. Styrene monomer is one of the materials used in the manufacture of glass fiber resin reinforced plastic. At present its annual output in Japan is estimated at about 1000000 tons. Recently the Japanese Ministry of Labor has decided to include styrene among 2nd class organic solvents in the Regulation of Prevention against Organic Solvent Poisoning. Since the response of animals to styrene monomer was reported by Spencer and co-workers in 1942⁷ there have been many reports on styrene poisoning especially in the recent past as reviewed by Ultramare and co-workers⁶. Since 1971 our department has carried out a series of studies on styrene poisoning with the objective to study the absorption, distribution and elimination of styrene in the living body which still require clarification^{1-5, 8-11}. Thus experiments have been carried out to assess the rate of uptake and elimination of styrene workers during the post-exposure period, and the distribution and elimination of styrene in rats exposed to styrene.

SUBJECTS, MATERIAL AND METHODS

Two studies were made on human beings, one on volunteers and another on styrene workers. The volunteers inhaled air containing styrene in concentrations of 10, 50 and 150 ppm in a Tefron bag for 10 minutes and the exhaled air was collected into another bag through a tube with a nonbreathing valve as shown in Figure 1. The rate of uptake was obtained from the following equation:

$$\text{rate of uptake (\%)} = (S_1 - S_2) / S_1 \times 100$$

where S_1 is the concentration of styrene in the inhaled air and S_2 that in the exhaled air.

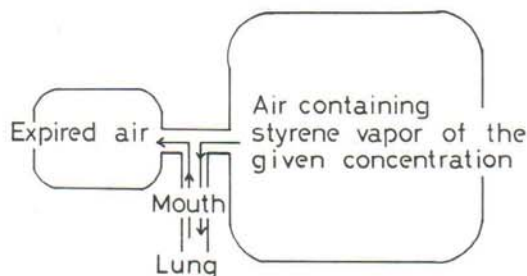


FIG. 1—Set-up for the experiment for the determination of styrene uptake.

In the study on styrene workers, the elimination of styrene during the post-exposure period was measured in three workers in a factory. They were working in atmospheric concentrations of styrene of about 80 ppm for about 5 hours; 3 hours in the morning and 2 hours in the afternoon. Immediately following the end of the day's work, the alveolar air was collected into a Tefron bag by the method of 20 seconds breath holding, and analyzed for styrene simultaneously with blood samples.

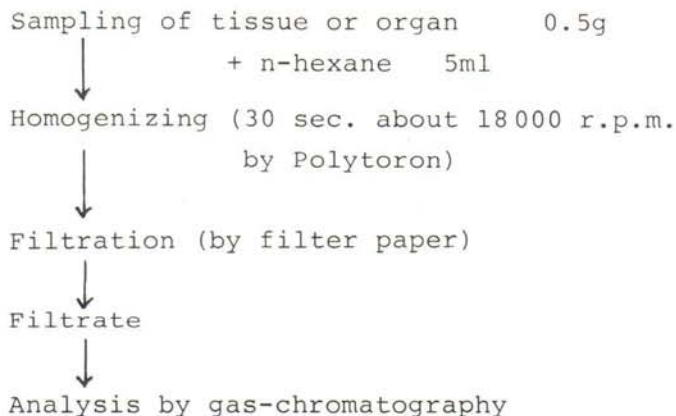


FIG. 2—Schematic diagram for analysis of styrene in tissues and organs.

In the animal experiment, rats were exposed to styrene vapor in various concentrations in exposure chambers under a controlled air flow rate of 2 l/min. After 4 hours of exposure to styrene, the blood, liver, kidney, spleen, brain, muscle and adipose tissue were excised from the ether-anesthetized rats. The excised tissues were washed with 0.85% saline, and wiped dry with filter paper. Then, 0.5 g of each tissue was taken into a tube with 5 ml n-hexane, homogenized by Polytoron and analyzed for styrene by gas-chromatography. Figure 2 shows the schematic diagram for the analysis of styrene in tissues and organs. Styrene in air samples of the exposure chambers was analyzed directly by means of gas-chromatography.

RESULTS AND DISCUSSION

Uptake of styrene in volunteers

The rate of uptake of styrene in humans exposed through inhalation in concentrations of 10, 50 and 150 ppm of styrene vapor was calculated at approximately 70% as shown in Table 1. Though the rate of uptake in female subjects seems to be greater than that in male subjects, there is no significant difference between the rates of uptake of both sexes.

TABLE 1
Uptake of styrene in volunteers (% of dose).

Concentration of styrene (ppm)	Men			Women		
	N	Mean	S.D.	N	Mean	S.D.
10	8	71.7	12.2	7	79.4	8.2
50	5	73.4	4.9	4	76.6	5.4
150	5	66.9	8.1	9	72.7	2.2

Elimination of styrene in styrene workers

A rapid elimination of styrene during the post-exposure period was observed in three workers exposed to about 80 ppm of styrene for about 5 hours as shown in Figures 3 and 4. The styrene concentrations in the alveolar air and blood during the post-exposure period were expressed as the following two phasic exponential equations:

$$C_{\text{alv}} = A \exp(-0.0039t) + B \exp(-0.046t)$$

$$C_{\text{b}} = A' \exp(-0.0039t) + B' \exp(-0.046t),$$

where C_{alv} is the styrene concentration in the alveolar air, C_{b} the styrene concentration in the blood, and A , B , A' and B' are the intercepts respectively. Since in these equations the rate constant for elimination in the alveolar air is almost equal to that for elimination in the blood, the styrene concentration in the blood can be derived from that in the alveolar air. The biological half times were estimated at about 40 minutes in the rapid phase, and at about 180 minutes in the slow phase, respectively.

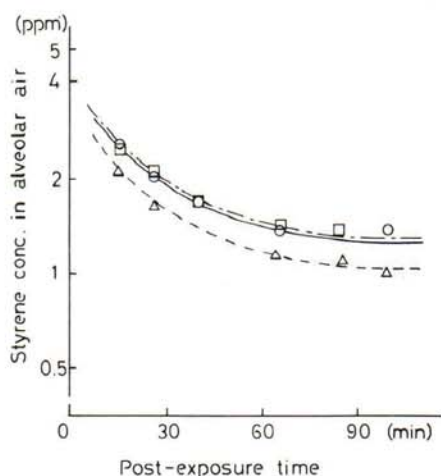


FIG. 3—Concentration of styrene in alveolar air of styrene workers during the post-exposure period. Circles = A; squares = B; triangles = C.

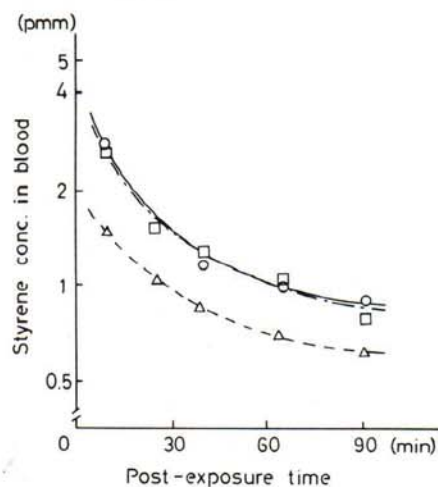


FIG. 4—Concentration of styrene in the blood of styrene workers during the post-exposure period. Circles = A; squares = B; triangles = C.

Determination of styrene in tissues

Before the animal experiment, the determination of styrene in tissues was performed. Already known amounts of styrene were added to the tissues, and the recovery rates were obtained with the following satisfactory results: 99.6% in the blood, 101.6% in the liver and 92.1% in the adipose tissue. Then, the variations of coefficients were obtained in rats following a 4-hour single exposure to styrene at 500 ppm. The results were as follows: 2.8% in the blood, 5.5% in the liver, and 14% in the adipose tissue.

Distribution and elimination of styrene in rats

The status of plateau was reached about 3 hours after exposure to styrene at about 500 ppm in rats as shown in Figure 5.

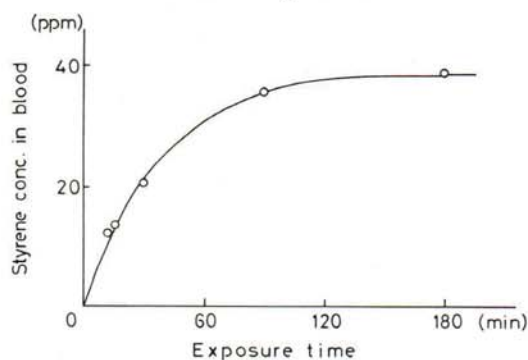


FIG. 5—Concentration of styrene in the blood of rats during exposure to styrene.

After a 4-hour single exposure to styrene at 500 to 1000 ppm in rats, the elimination of styrene from the blood, liver, kidney, spleen, brain and muscle was expressed by the following equation:

$$C = A \exp (-\alpha t) \text{ ————— (1)}$$

where C is the concentration of styrene in the organs and tissues at time t, α the rate constant for elimination, and A the intercept. Figure 6 shows the elimination of styrene from the blood as a representative of these tissues.

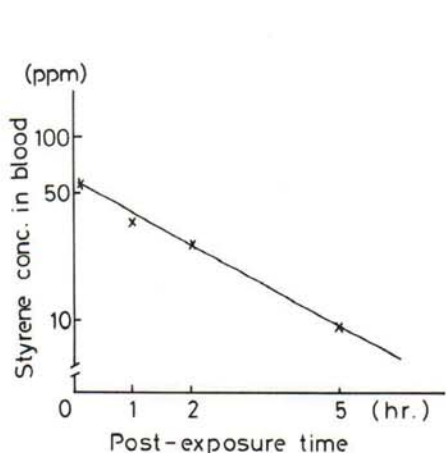


FIG. 6—Concentration of styrene in the blood of rats after a single exposure to about 700 ppm styrene.

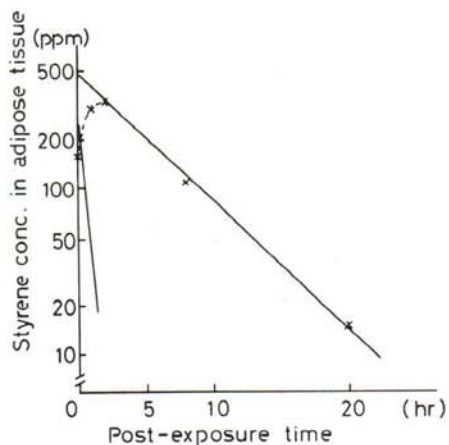


FIG. 7—Concentration of styrene in the adipose tissue of rats after a single exposure to about 700 ppm styrene.

As shown in Figure 7, the elimination from adipose tissue was expressed by the following equation:

$$C = P \exp (-\alpha_1 t) - Q \exp (-\alpha_2 t) \text{ ————— (2)}$$

where C is the concentration of styrene in adipose tissue, α_1 the rate constant for elimination, and α_2 the rate constant for absorption, while P and Q are the intercepts.

The biological half time (BHT) was estimated from the following equation:

$$t = 0.693/\alpha \text{ or } t = 0.693/\alpha_1.$$

The BHTs were about 2 hours for the organs and tissues except adipose tissue, and about 6 hours for adipose tissue.

There were no significant differences in the rate constant for elimination and BHT inside and between the organs and tissues, except adipose tissue.

When $t = 0$ in equations 1 and 2, the distribution of styrene in the organs and tissues was obtained. The relative ratio of the distribution of styrene in the organs and tissues, when styrene concentration in the blood was 1, was in decreasing order as follows: adipose tissue >> liver > brain > kidney > blood = spleen > muscle (see Table 2).

TABLE 2
Pharmacokinetic studies of styrene in tissues and organs following a single exposure to styrene at 500 to 1000 ppm in rats.

		Blood	Liver	Kidney	Spleen	Muscle	Brain	Adipose tissue
Rate constant of elimination	Mean	0.30	0.33	0.34	0.33	0.37	0.35	0.11
	S.D.	0.075	0.092	0.097	0.096	0.13	0.11	0.065
Biological half time (h)	Mean	2.4	2.1	2.2	2.2	2.0	2.1	6.3
	S.D.	0.56	0.51	0.49	0.53	0.58	0.55	4.5
Distribution	Mean	1	2.8	1.5	0.93	0.68	2.1	35
	S.D.		0.71	0.19	0.13	0.18	0.36	22

Almost the same results were obtained in an experiment with intraperitoneal administration of styrene. As shown in Figure 8, repeated 4-hour exposures at about 700 ppm per day for five days led to a distribution and elimination of styrene in organs and tissues similar to those in a single exposure. No accumulation of styrene in the body was observed.

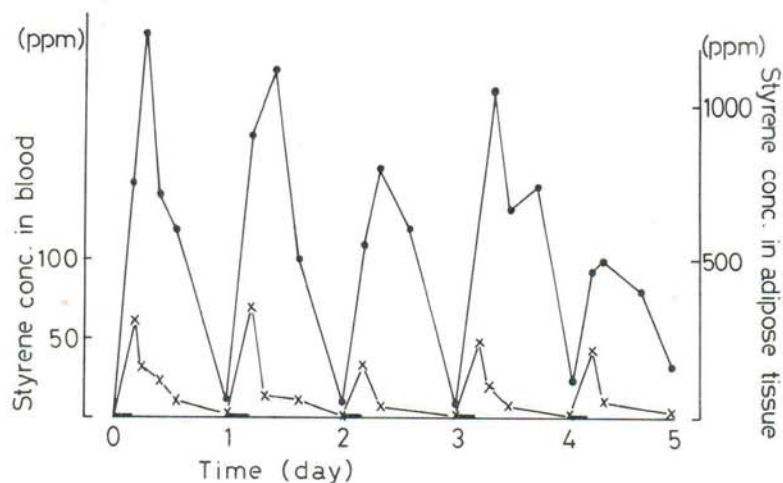


FIG. 8 - Variation of styrene in blood and adipose tissue after repeated exposures to about 700 ppm styrene. Crosses = blood; solid circles = adipose tissue; thick lines = styrene exposure of 4 hours.

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