

Distribution and elimination of ^{14}C -xylene in rat

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CARLSSON A. Distribution and elimination of ^{14}C -xylene in rat. *Scand j work environ health* 7 (1981) 51—55. Male rats were exposed to about 45 ppm of radioactively labeled xylene in the inspired air for 1—8 h. The distribution and elimination of xylene and metabolites in different organs and tissues were studied. To get an idea of the ratio between unmetabolized and metabolized xylene in organs and tissues, a supplementary study was performed in which five male rats were exposed to about 250 ppm of radioactively labeled xylene. The concentration of xylene and metabolites in the subcutaneous fat, where the largest amounts were found, displayed a sharp rise during the first 4 h of exposure in comparison with the following 4 h. Both during exposure and the decay phase the concentration of xylene and its metabolites in the cerebrum, cerebellum and muscles paralleled the arterial blood concentration and amounted to about 40 % of the arterial blood value for xylene. The coefficient between the concentration of unmetabolized xylene in subcutaneous fat and inspiratory air was calculated to be 525 after 8 h of exposure.

Key terms: exposure, metabolites, tissue concentration, xylene.

The amount of xylene exhaled in an unchanged form by man via the lungs following exposure is about 5 % (1, 8).

About 95 % of the uptake of p-xylene in man is excreted as methylhippuric acid (8). The main metabolite in rat is also methylhippuric acid (7).

In the present study rats were exposed to p-xylene in the inspiratory air. The objective was to study the distribution and elimination of p-xylene and its metabolites.

Experimental design and methods

Male rats (Sprague-Dawley, 200 g) were exposed to a mixture of xylene vapor and air for 1—8 h. The experiments were performed in the same manner as in a similar study on styrene (4). Radioactively labeled p-xylene (methyl- ^{14}C) with a specific activity of 0.35 mCi/mmol was used, and the

concentration of xylene in the inspiratory air was 208 ± 13 (SD) mg/m³ (47.8 ppm). After the conclusion of the exposure some rats breathed ordinary air during a decay phase. Table 1 lists the exposure durations, decay phases, and the number of rats exposed to xylene.

After the rats had been killed by luxation of the cervical spine, double samples were taken from the liver, kidneys, subcutaneous fat, cerebrum, cerebellum, muscles, lungs, spleen, ischiadic nerve, and arterial blood for the assay of radioactively labeled solvent and radioactive metabolites according to the methods described in the styrene study (4). The results have been determined on the basis of the mean value of the two samples. The error of the method was calculated for each organ and tissue according to the method for double determinations with systematic difference.

To get an idea of the ratio between the unmetabolized and metabolized xylene in the organs and tissues, a supplementary study was performed. A total of five male rats (Sprague-Dawley, 200 g) were utilized. The animals were exposed to radioactively labeled p-xylene (methyl- ^{14}C) in the in-

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Table 1. Organ and tissue concentrations (nmol/g tissue) of xylene equivalents (xylene and its metabolites) in rat following exposure to a xylene concentration of 208 mg/m³ (47.8 ppm) in inspiratory air — Means $\pm$ standard errors (determined from the mean of two samples for each rat) and the method error.

Organ	Duration of exposure + time after exposure (h)									Method error ^a (% of mean value)
	1 + 0 (N = 4)	2 + 0 (N = 4)	4 + 0 (N = 4)	8 + 0 (N = 4)	1 + 1 (N = 4)	1 + 3 (N = 4)	1 + 6 (N = 4)	8 + 1 (N = 4)	8 + 3 (N = 4)	
Liver	23 $\pm$ 2	22 $\pm$ 1	27 $\pm$ 2	37 $\pm$ 4	4 $\pm$ 1	1 $\pm$ 0	1 $\pm$ 0	10 $\pm$ 2	9 $\pm$ 4	2.3
Kidneys	527 $\pm$ 100	497 $\pm$ 139	1,080 $\pm$ 366	719 $\pm$ 131	250 $\pm$ 43	44 $\pm$ 7	17 $\pm$ 7	557 $\pm$ 130	475 $\pm$ 160	24.3
Subcutaneous fat	76 $\pm$ 8	125 $\pm$ 14	182 $\pm$ 6	270 $\pm$ 7	61 $\pm$ 5	38 $\pm$ 2	11 $\pm$ 2	260 $\pm$ 20	202 $\pm$ 14	9.6
Cerebrum	10 $\pm$ 2	10 $\pm$ 1	11 $\pm$ 1	12 $\pm$ 1	2 $\pm$ 1	1 $\pm$ 0	0	4 $\pm$ 0	4 $\pm$ 1	6.6
Cerebellum	9 $\pm$ 0	10 $\pm$ 1	11 $\pm$ 2	12 $\pm$ 1	2 $\pm$ 1	1 $\pm$ 0	0	4 $\pm$ 1	3 $\pm$ 0	7.6
Ischiadic nerve	35 $\pm$ 7	51 $\pm$ 9	63 $\pm$ 14	64 $\pm$ 10	24 $\pm$ 3	10 $\pm$ 2	2 $\pm$ 0	35 $\pm$ 4	16 $\pm$ 3	—
Muscles	10 $\pm$ 2	10 $\pm$ 0	11 $\pm$ 2	13 $\pm$ 3	4 $\pm$ 1	1 $\pm$ 0	1 $\pm$ 0	9 $\pm$ 1	8 $\pm$ 1	3.7
Lungs	21 $\pm$ 2	21 $\pm$ 5	21 $\pm$ 1	24 $\pm$ 3	4 $\pm$ 1	1 $\pm$ 0	0	8 $\pm$ 1	8 $\pm$ 3	5.2
Spleen	12 $\pm$ 2	10 $\pm$ 1	8 $\pm$ 1	10 $\pm$ 1	2 $\pm$ 1	0	0	4 $\pm$ 1	2 $\pm$ 1	9.0
Blood	25 $\pm$ 4	23 $\pm$ 2	28 $\pm$ 3	30 $\pm$ 2	5 $\pm$ 1	2 $\pm$ 0	1 $\pm$ 0	8 $\pm$ 2	4 $\pm$ 1	4.1

a SD = $\{[\sum D^2 - (\sum D)^2/n]/2(n-1)\}^{1/2}$.

Table 2. Mean organ and tissue concentrations of xylene (X) and its metabolites (M), specified as nanomoles of xylene equivalents per gram of tissue, in rat after exposure to a xylene concentration of 1,117 mg/m³ (257 ppm) in inspiratory air, the proportion of metabolites as percentage, and the error of the method for xylene.

	Duration of exposure + time after exposure (h)										Method error for xylene ^a (% of mean value)
	0.25 + 0 (N = 1)		1 + 0 (N = 1)		4 + 0 (N = 1)		8 + 0 (N = 1)		8 + 3 (N = 1)		
	X	M	X	M	X	M	X	M	X	M	
Liver nmol/g	—	—	89	104	80	160	283	283	5	113	7.8
%				54		67		50		95	
Subcutaneous fat nmol/g	—	—	894	264	2,147	895	5,529	1,846	3,193	867	8.1
%				.23		29		25		21	
Muscles nmol/g	—	—	19	47	24	52	57	89	14	9	11.9
%				71		69		61		40	
Cerebrum nmol/g	—	—	80	57	71	57	254	57	14	5	9.4
%				41		44		18		25	
Blood nmol/g	33	80	47	104	61	104	113	170	5	89	5.6
%		71		69		63		60		95	

a SD = $\{[\sum D_{12}^2 - (\sum D_{12})^2/n] + [\sum D_{23}^2 - (\sum D_{23})^2/n] + [\sum D_{13}^2 - (\sum D_{13})^2/n]\}^{1/2}/[6(n-1)]^{1/2}$.

spiratory air for 0.25–8 h with one rat at each exposure time (table 2). The specific activity for p-xylene was 0.07 mCi/mmol, and the air mixture contained $1,117 \pm 78$ (SD) mg/m³ (257 ppm). Samples were isolated from the liver, subcutaneous fat, muscles, cerebrum, and arterial blood for the analysis of unmetabolized and metabolized p-xylene. The experimental and analytical methods have been described in the supplementary styrene study (4). Three samples were taken from each organ and tissue, and the results are listed as the mean value of the three samples. The error of the method was calculated for each organ and tissue according to the method for triple determinations with systematic difference.

Results

The concentrations of p-xylene and its metabolites (xylene equivalents) are shown in fig 1. The subcutaneous fat displayed a sharp rise in xylene equivalents with increasing exposure duration. After 4 h of exposure, the subcutaneous fat contained 182 nmol of xylene equivalents/g of tissue. Following 8 h of exposure, the concentration had increased to 270 nmol/g. After 8 h of exposure, the corresponding concentration amounted to 64 for the ischiadic nerve, 37 for the liver, and to 12 for the cerebrum. The concentration of xylene equivalents in the cerebrum, cerebellum, muscles, spleen and lungs paralleled that of the arterial blood throughout the entire

period of exposure. It was about 80 and 40 % of the blood value for the lungs and other organs, respectively. The arterial blood contained 30 nmol of xylene equivalents/g of tissue after 8 h of exposure. The concentration of xylene equivalents in the kidneys was high (about 700 nmol/g of tissue) throughout the entire period of exposure.

In fig 2 the organ and tissue concentrations during a 6-h decay period are shown. The exposure to p-xylene lasted 1 h. After a 6-h decay phase the subcutaneous fat contained about 15 % of the initial value. The concentrations in the cerebrum, cerebellum, and muscles paralleled the blood concentration also during the decay phase and amounted to about 40 % of the blood value (table 1, fig 2). In the liver the concentration was very low during the decay phase.

The half-time for the xylene equivalents in the subcutaneous fat was estimated to be 2.2 h after 1 h of exposure and 6.9 h after 8 h of exposure. The accumulation of xylene equivalents in different organs and tissues is listed completely in table 1.

In fig 3 the relative concentration of metabolized xylene in relation to the total quantity of xylene and its metabolites, ie, xylene equivalents, is shown (supplementary study). During the period of exposure the liver contained 50 to 67 % metabolites. Three hours after the exposure was concluded, the proportion increased to about 95 %. The percentage of metabolites in the muscles ranged from 61 to 71 % during exposure. After the 3-h decay period it decreased to about 40 %. The percentage of metabolites in the subcutaneous fat was relatively constant both during and after exposure and averaged

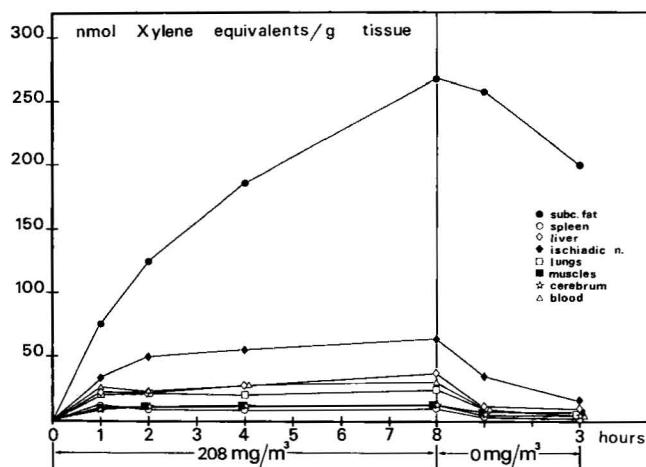


Fig 1. Organ and tissue concentrations of xylene equivalents (xylene and its metabolites) in rat immediately after the conclusion of exposure as a function of the exposure duration in hours. The animals were exposed to a xylene concentration of 208 mg/m³ (47.8 ppm). (subc. = subcutaneous, n. = nerve)

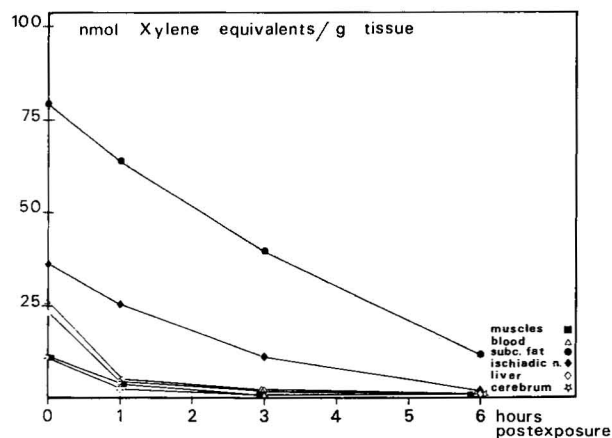


Fig 2. Organ and tissue concentrations of xylene equivalents (xylene and its metabolites) in rat as a function of the time elapsed after the conclusion of exposure. The animals were exposed to a xylene concentration of 208 mg/m³ (47.8 ppm) for 1 h. (subc. = subcutaneous, n. = nerve)

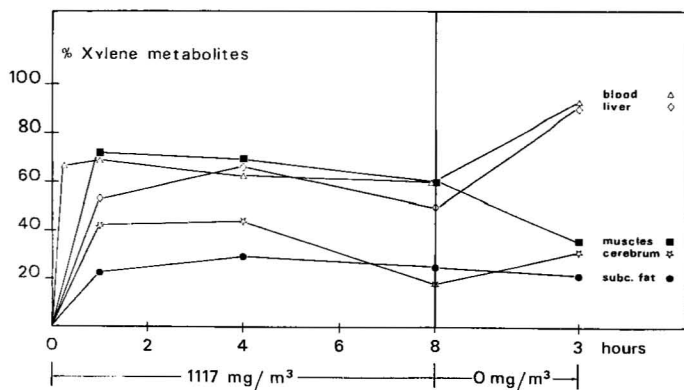


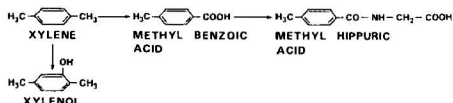
Fig 3. The concentration of metabolized xylene in rat, expressed as a percentage of the total quantity of xylene and its metabolites in different organs and tissues during an exposure lasting up to 8 h, followed by a 3-h decay period. The animals were exposed to 1,117 mg/m³ (257 ppm) of xylene. (subc. = subcutaneous)

about 25 %. During exposure the arterial blood contained 60–70 % metabolites. Three hours after the conclusion of exposure the concentration increased to about 95 %.

The quotients between the concentrations of unmetabolized xylene in subcutaneous fat and the concentration in blood or air were calculated. After 8 h of exposure the quotient was about 50 for subcutaneous fat/blood and 525 for subcutaneous fat/air. The accumulation of xylene and metabolites is listed completely in table 2.

Discussion

Xylene is found in three different isomeric forms, ie, ortho-, meta- and para-xylene, with slightly varying metabolism in animals (3). Para-xylene is metabolized according to the following diagram (7):



A remarkable finding in the present xylene study was the proportionately low concentrations of xylene equivalents in organs and tissues in comparison to the corresponding equivalents of styrene in another study (4). After 8 h of exposure the concentration of xylene equivalents in the arterial blood was only about 60 % of the corresponding value in the styrene study, in spite of a somewhat higher xylene concentration in the inspiratory air. In exposure of man to about 650 mg/m³

(150 ppm) of styrene for 2 h during work with increasing work loads (2), the coefficient between the concentration of styrene in arterial blood and inspiratory air was about 25. In a corresponding exposure of man to about 435 mg/m³ (100 ppm) of xylene (1), the coefficient was about 16. These findings suggest a higher coefficient between arterial blood and inspiratory air for styrene than for xylene in exposure of animals and man in vivo. The lower concentrations of xylene equivalents in the other organs and tissues, compared to those of styrene (4), depend on the low concentration of xylene equivalents in the arterial blood, which transports the solvent to the tissues.

After 2 h of exposure to approximately 50 ppm of xylene in the present study, a concentration of about 122 nmol of xylene equivalents/g of tissue was found in the subcutaneous fat. In exposure of man to 100 ppm of xylene for 2 h during work with increasing work loads (5), the concentration of xylene in subcutaneous fat amounted to only 38 nmol/g after the conclusion of exposure. In the present study the half-time for xylene equivalents in the subcutaneous fat after 1 h of exposure was estimated to be 2.2 h. Engström et al (5) showed that the elimination of xylene from adipose tissue in man is slow, with a mean concentration 22 h after exposure on about the same level as during the first 4 h after exposure. Engström & Riihimäki (6) estimated the half-time for xylene in the subcutaneous fat of man to be about 58 h. The reasons for a faster distribution and elimination in rat than in man has been discussed in another report (4).

From the supplementary study the coefficient between the concentration of un-metabolized xylene in subcutaneous fat and inspiratory air was calculated after 4 and 8 h of exposure. The coefficient was 204 after 4 h and 525 after 8 h of exposure. In vitro experiments (9) showed an oil/air partition coefficient of 4,050 for xylene. The reasons for different values from experiments in vivo and in vitro have also been discussed in another report (4).

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