CHANGES IN CONTENT OF ASCORBIC ACID AND NUCLEIC ACIDS PRODUCED BY BENZENE AND FORMALDEHYDE

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Inhalation of benzene and formaldehyde vapors changes the content of ascorbic acid and nucleic acids in certain organs of female rats and their fetuses. This indicates the possible teratogenic action of the substances tested.

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The object of this investigation was to study the possible teratogenic action of microconcentrations of benzene and formaldehyde when inhaled experimentally by rats. To assess the reaction of the female rat and her fetus the content of ascorbic acid (AA) and of nucleic acids (RNA and DNA) was determined in the fetuses, placenta, and the liver of the mother rat and fetus.

We used determination of RNA and DNA in the organs because of the dominant role of nucleic acids in protein synthesis, in the transmission of inherited characteristics, and in many other metabolic processes. Changes in AA metabolism are an index of the harmful effect of chemical substances [2, 6, 8].

EXPERIMENTAL METHOD

Experiments were carried out on 160 female albino rats and 1294 fetuses. The action of inhalation of different concentrations of benzene (670, 63.3, 56.6, 20.4, 5.6, and 1 mg/m^3) and formaldehyde (1 and 0.012 mg/m^3) throughout the 24-h period was studied. Between 5 and 12 females were placed at a time in a chamber containing a definite concentration of the test substance. After 20 days the females were mated for 6-10 days with males which had not been poisoned, in the proportion of one male to 3-4 females. The females remained in the chamber until parturition, and continued to be exposed to the action of the test substances. For each series of experimental animals there was a corresponding group of controls which were kept in identical conditions (food, temperature, humidity, velocity of air movement), except that pure air was supplied to the chamber.

After the end of the experiment the females were sacrificed by decapitation and the content of AA and nucleic acids in their organs determined by the usual methods [5, 7]. The results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

The results obtained are given in Tables 1 and 2. No visible developmental fetal malformations were found in any series of experiments. With an increase in benzene concentration to 670 mg/m^3 the number of fetuses per female tended to zero (13.3 in the control group). The number of fetuses expressed per female also decreased after exposure to formaldehyde (from 11.3 in the control group to 9.8 after the action of 0.012 mg/m^3 and 8.6 after the action of 1 mg/m^3). As a rule, following exposure to benzene the weight of the fetuses decreased and the relative weights of their organs were reduced. Both concentrations of formaldehyde caused an increase in weight of the fetuses and of their organs, possibly as a result of the embryotropic action of these substances.

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TABLE 1. Content of Ascorbic Acid in Whole Fetus, Placenta, and Liver of Mother Rats and Fetuses Exposed to Different Concentrations of Benzene, M ± m

Concentration of benzene (in mg/m³)	No. of animals	AA content (in mg%)			
		whole fetus	placenta	maternal liver	fetal liver
63,3	5	18,0± 1,2	12,7± 0,8	6,8± 0,05[3]	16,7± 1,5[1]
Control	5	22,5± 2,2	14,9± 3,0	28,7± 1,4	20,9± 0,4
56,6	5	20,3± 1,8[1]	9,9± 0,5[3]	19,4± 1,0[2]	17,0± 0,6[3]
20,4	10	25,7± 0,9	16,5± 1,1	19,4± 1,1[2]	—
5,6	6	27,7± 1,1	12,3± 0,8[1]	22,9± 2,1	22,1± 0,5
1,0	8	24,6± 0,8	20,7± 1, 0[2]	23,5± 0,7[2]	21,0± 0,4
Control	5	25,2± 0,5	16,9± 0,04	31,2± 0,4	22,4± 1,1

Note. Here and in Table2: [1] P = 0.05, [2]P = 0.01, [3]P = 0.001.

TABLE 2. Content of Ascorbic Acid in Whole Fetus, Placenta, and Liver of Mother Rats and Fetuses Exposed to Formaldehyde, M ± m.

Concentration of formaldehyde (in mg/m³)	No. of animals	AA content (in mg %)			
		whole fetus	placenta	maternal liver	fetal liver
1,0	10	14,3± 0,7[2]	6,4± 0,7	16,8± 1,1[1]	14,8± 0,7
0,012	10	14,4± 0,2[3]	6,7± 0,7	18,1± 2,1[2]	20,1± 1,7[1]
Control	10	19,0± 1,1	9,7± 1,4	20,6± 0,8	15,8± 0,4

Table 1 gives data for the AA content in the whole fetus, the placenta, and the liver of the mother rats and fetuses of the control group of animals and females exposed to different concentrations of benzene. These results in Table 1 show that after inhalation of benzene the AA content in the tested organs fell, and the higher the concentration of benzene the greater the decrease in AA content. The AA content fell first of all in the maternal liver and later in the placenta and fetal liver.

In Table 2 the results of investigation of the AA content in the organs after exposure to formaldehyde are given. A statistically significant decrease in the AA content in the maternal liver and the whole fetus was found.

The results indicating a decrease in AA content in the maternal organs are in complete agreement with those in the literature [4, 10], according to which the fetal blood during pregnancy contains twice as much vitamin C as the maternal blood, while the AA content in the liver of the rabbit embryo is 1.5 times greater than in the maternal liver. The important role of the placenta in supplying vitamin C to the fetus should also be mentioned [1, 3]. This special function of the placenta in supplying AA to the fetus is specific for this vitamin alone, for experiments with vitamins A and B_1 did not yield similar results [9].

The decrease in AA content in the liver and placenta was evidently due to the fact that the mother rats attempted to create optimal conditions for fetal development. During exposure to small concentrations of benzene and formaldehyde, the mother was able to compensate for the AA deficiency in the fetus; during exposure to high concentrations, despite mobilization of all defensive and adaptive mechanisms, the mother was unable to compensate for the fetal AA deficiency. This is confirmed by the decrease in AA content in the whole fetus following exposure to both concentrations of formaldehyde (Table 2).

These results are in agreement with observations on human subjects. In a group of workers of the ZIL omnibus factory with occupational benzene poisoning resulting in ovarian hypofunction, we found a decrease in the blood AA concentration (0.36 ±0.02 mg%) compared with clinically healthy female workers in the same factory (0.49 ±0.03 mg %; P < 0.001). The last series of experiments showed that formaldehyde lowers the DNA content and increases the RNA content in the organs. Conversely, following exposure to benzene in concentrations of between 1 and 56.6 mg/m³, the RNA content in the placenta and in the fetal liver and brain decreased while the DNA content increased. In the maternal liver the DNA content fell with an increase in the benzene concentration.

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