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PLACENTAL TRANSFER OF SULFAMETHAZINE IN MICE AND RATS

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Abstract

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Placental transfer of sulfamethazine in mice was investigated in the second half of their pregnancy. The transfer was not free but only a part of the drug administered to mothers was found to pass from the maternal circulation to the fetus.

Placental transfer of sulfamethazine in rats was investigated at two stages of gestation – early (8th to 10th day) and late (18th to 20th day of gestation). There was free and rapid passage of the drug from the maternal circulation to the fetus at the early stage of gestation. But the passage was found to be greatly reduced at the late stage of gestation. The drug also appeared in the amniotic fluid.

Mice, rat, placenta, fetus, transfer, sulfamethazine.

The placenta in unique is its function. Its life time is short relative to the fetus, and its size and function change continually during the course of gestation (Wynn 1968). The study of the placental transfer of drugs is of paramount importance because much of intrauterine fetal welfare depends on the exchange processes of these substances through the placenta.

Sulfonamides have been reported to easily pass through both human and animal placenta (Lee, Anderson and Chen 1938; Friesen 1951; Siddique and Šimůnek 1977). The possible use of these drugs for intrauterine therapy as well as for fetal prophylaxis and their effects on the fetus and newborn make it necessary to know and understand the degree and extent of transplacental passage of these drugs.

The present paper reports on the placental transfer of sulfamethazine in mice in their second half of pregnancy and in rats at two stages of pregnancy.

Materials and Methods

White mice and Wistar rats were obtained from the Biological Research Center, Konárovice near Prague. Sulfamethazine used was a product of Czechoslovak national pharmaceutical enterprise, Spofa.

Procedure for mice

13 mice which were 15 to 18 days pregnant were divided into two groups and injected intramuscularly a freshly prepared 2 % solution of sodium-sulfamethazine at the dose of 0.2 g/kg body mass. One group of mice was killed after 2 hours and the other group after 24 hours of drug administration. Samples of blood, placenta and fetus of each group of mice were collected and their sulfamethazine concentrations measured. In all there were 70 fetal samples. Simultaneous control experiment with 5 nonpregnant adult female mice was carried out.

Procedure for rats

Two independent experiments were done. In both cases, transfer was studied at two stages of gestation.

Experiment 1:

22 rats were on the 8 th to 10 th day of pregnancy and 15 on the 18th to 20th day of pregnancy. At each of these two stages of pregnancy the rats were divided into 4 groups and injected intravenously a freshly prepared 10 % sodium-sulfamethazine solution at the dose of 0.15 g/kg body mass. Rats were decapitated and samples of their blood and fetuses collected at intervals of 10, 30, 60 and 120 minutes from the time of drug administration; one group being decapitated at each of the time periods. Placental samples of only 18 to 20 days pregnant rats were collected because placentas of 8 to 10 days pregnant rats could not be separated from their fetuses. Simultaneous control experiments with 15 nonpregnant adult female rats were done on each occasion.

Experiment 2:

Placental transfer of sulfamethazine was investigated on the 10th day and on the 20th day of gestation. On each of the 10th and 20th day the rats were divided into 4 groups with at least 6 pregnant rats in each group. They were injected intramuscularly a freshly prepared 10 % sodium-sulfamethazine solution at the dose of 0.15 g/kg body mass and were killed after 15, 60, 120 and 240 minutes of drug administration; one group of rats being killed at each time period. Rest of the procedure was same as in the case of Experiment 1.

Sulfamethazine concetrations of all samples of mice and rats were determined according to the method of Bratton and Marshall (1939), modified by Wagner (1950). The mean, S. E. and level of confidence were determined according to Czechoslovak Pharmacopoea (Ph Bs-3, 1970). Mean sulfamethazine level was expressed as mg/100 ml.

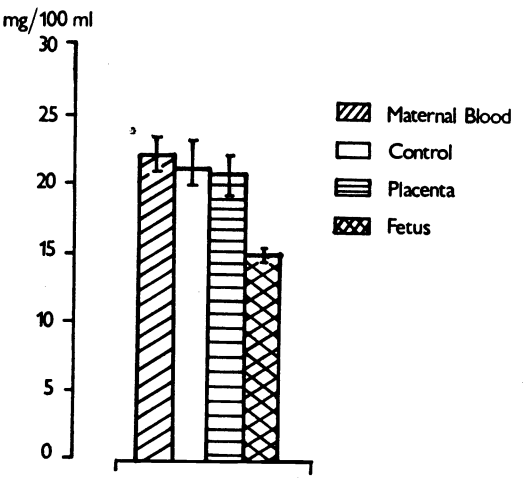

Fig. 1.
Sulfamethazine levels (mg/100 ml) in blood, placentas and fetuses of pregnant mice and in blood of nonpregnant control mice after 2 hours of intramuscular administration of a 2 % sodium-sulfamethazine solution at the dose of 0.2 g/kg body mass.

Results

Mice experiments

Results including the blood levels of nonpregnant control mice are illustrated in Fig. 1. There was no significant difference of blood levels of pregnant mice from those of nonpregnant ones. Sulfamethazine levels of 25 placentas from 9 mice, out of the total of 70 placentas from 13 mice, were higher than those of the corresponding maternal blood. Sulfamethazine levels of rest of the placental samples were lower than those of the maternal blood. However, the difference between the mean maternal blood level ($24.57 \pm \pm 2.03$ mg/100 ml) and the mean placental level (22.95 ± 1.18 mg/100 ml) was not statistically significant (for 90 % possibility or even for 50 % possibility) as was determined by t-test according to Myslivec (1957). Fetal levels were always found to be considerably lower than both maternal blood and placental levels. Highest and lowest fetal levels were 84 % and 48 % respectively of the corresponding maternal blood level (= 100 %).

Rat experiments

Experiment 1.

Results including the blood levels of nonpregnant control rats are shown in Fig. 2. Usually the blood levels of 8 to 10 days pregnant rats were lower than those of the 18 to 20 days pregnant ones.

As is evident from the graph, there appeared to be no significant barrier to the passage of the drug from the maternal circulation to the fetus on the 8th to 10th day of pregnancy.

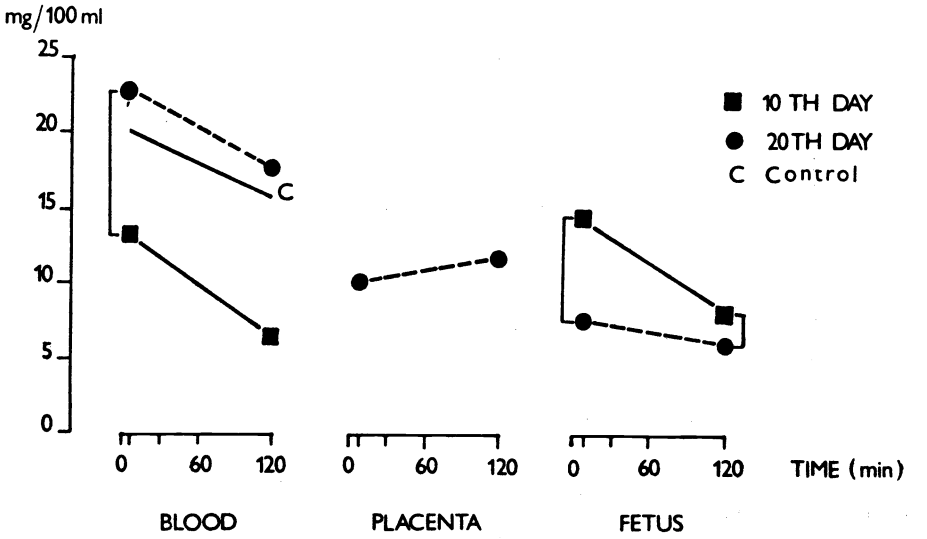

Fig. 2.

Sulfamethazine levels (mg/100 ml) at different time periods after i. v. administration of a 10 % sodium-sulfamethazine solution to pregnant (on the 8—10 day and on the 18—20 day of pregnancy) and nonpregnant (control) rats at the dose of 0.15 g/kg body mass. (Blood levels of pregnant rats and their placental and fetal levels after 10 and 120 minutes of drug administration are connected by lines.)

However, on the 18th to 20th day of gestation only a part of the total sulfamethazine administered to mothers could be found to pass through the placenta to their fetuses. Both placental and fetal levels were lower than the corresponding maternal blood levels and the absolute fetal level was always found to be lower than the corresponding placental level. Both maternal blood and fetal levels were found to fall as time elapsed after drug administration. But the placental level was found to rise even after 120 minutes of drug administration.

Experiment 2.

10th day of gestation:

Data including the blood levels of nonpregnant control rats are shown in Fig. 3. After 15 minutes of drug administration, the maternal blood level was 11.40 mg/100 ml while the corresponding fetal level was 6.62 mg/100 ml which represented about 58 % of the maternal blood level. After 60 minutes, the maternal blood level rose to 15 mg/100 ml and the fetal level to 12.18 mg/100ml. After 120 minutes of drug administration, the maternal blood level further rose to 18.73 mg/100 ml and the fetal level to 15.45 mg/100 ml which represented about 82 % of the maternal blood level at that time period. Both maternal blood and fetal levels were found to have considerably dropped after 240 minutes of drug admini-

stration. The maternal blood level dropped to 15.81 mg/100 ml, and the fetal level to 14.44 mg/100 ml and this was about 92 % of the matenal blood level at that time period.

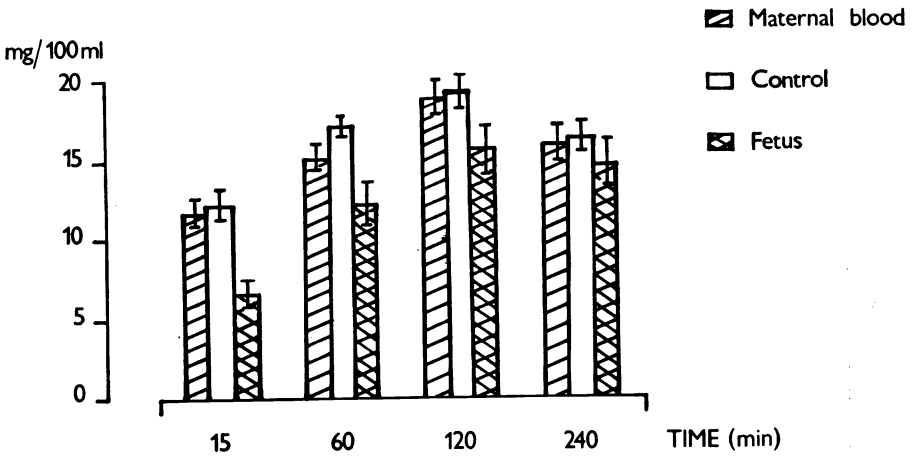

Fig. 3.
Sulfamethazine levels (mg/100 ml) at different time periods after i. m. administration of a 10 % sodium-sulfamethazine solution to pregnant rats on the 10th day of pregnancy and to nonpregnant (control) rats at the dose of 0.15 g/kg body mass.

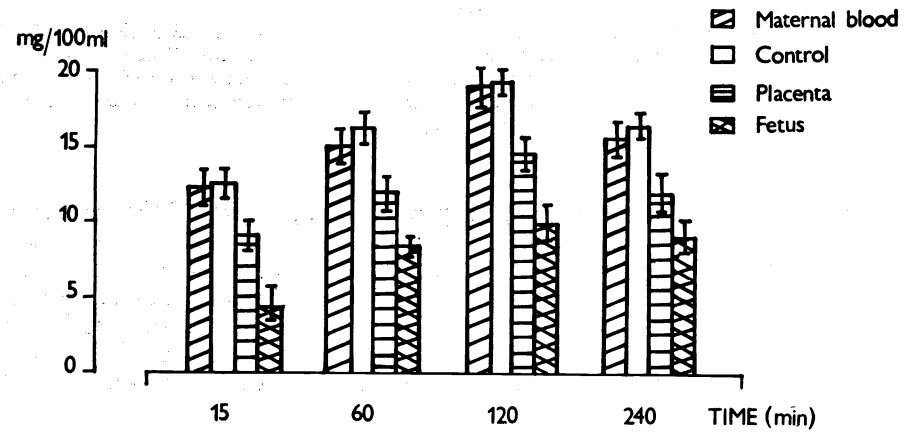

Fig. 4.
Sulfamethazine levels (mg/100 ml) at different time periods after i. m. administration of a 10 % sodium-sulfamethazine solution to pregnant rats on the day 20 of gestation and to nonpregnant (control) rats at the dose of 0.15 g/kg body mass.

20th day of gestation:

Results including the blood levels of nonpregnant control rats are illustrated in Fig. 4. After 15 minutes of drug administration, the maternal blood level was 12.15 mg/100 ml, and the fetal level was 4.33 mg/100 ml which was about 36 % of the maternal blood level. After 60 minutes, the fetal level was 8.42 mg/100 ml while the maternal blood level was 15.17 mg/100 ml. After 120 minutes, the mater-

nal blood level rose to 19.15 mg/100 ml and the corresponding fetal level to 10.23 mg/100 ml which was about 53 % of the maternal blood level. After 240 minutes of drug administration, the maternal blood level dropped to 15.59 mg/100 ml while the fetal level dropped to 9.13 mg/100 ml which amounted to about 59 % of the maternal blood level.

The placental level was always found to be lower than the corresponding maternal blood level but higher than the corresponding fetal level. After 15 minutes of drug administration, the placental level was found to be 9.49 mg/100 ml. The highest placental level of 14.66 mg/100 ml which was reached after 120 minutes of drug administration was about 76 % of the corresponding maternal blood level. The drug also appeared in amniotic fluid. After 15 minutes, the amniotic fluid level was 2.36 mg/100 ml. The level further rose to 6 mg/100 ml and 7.37 mg/100 ml after 60 and 120 minutes respectively. Even after 240 minutes the amniotic fluid level did not drop but was found to rise slowly to 7.55 mg/100 ml.

Discussion

Since the first observation on the placental transfer of sulfanilamide in rabbit by Lee et al. (1938), it has been repeatedly shown that all commonly used sulfonamides readily cross the placenta into the fetal circulation, in some cases within 15 minutes of intravenous administration, equilibrium being reached with maternal blood level in approximately 3 hours with the fetal level about 10 to 30 % below that in the mother (Speert 1938, 1940; Anderson and Simesen 1942; Friesen 1951). However, the stage of gestation has been suggested to be a factor of great importance in placental transfer (Snyder and Speert 1938). Different studies made in our laboratory have demonstrated that the stage of gestation and the species of animals are factors of great importance in placental transfer processes (Kubíková, Lenhartová and Hegerová 1976; Siddique and Šimůnek 1977, 1978).

The present report is in agreement with other reports on the placental transfer of sulfonamides in animals and it particularly confirms that the stage of gestation is a great factor in placental transfer. Our present data on mice show that there was no free transplacental passage of sulfamethazine on the 15th to 18th day of pregnancy indicating the presence of a relative barrier to the transfer of this drug through the placenta of mice at this stage of gestation. Our present data on rats show significant difference in placental permeability to sulfamethazine at different stages of gestation. There was free and rapid transfer of the drug on the 10th day of gestation. Equilibrium between the maternal blood level and the fetal level appeared to have reached 2—4 hours after drug administration. But this transfer became greatly reduced on the 20th day of gestation indicating the presence, and as such, gradual emergence of a relative placental barrier to sulfamethazine with the advancement of pregnancy. This apparent difference in permeability to sulfamethazine at different stages of gestation may be due to morphological or physiological alterations or limitations of the placenta during the course of gestation in rats.

There is considerable variation in membrane thickness within an individual placenta as well as significant change is placental histology as gestation proceeds (Wynn 1968). Number of effective placental layers vary in the same species from week to week even in the corresponding portion of the same placenta (Longo 1972). Kiryushenkov et al. (1971) reported that the placental permeability to oxacillin decreased as gestation progressed in rat.

Regarding the appearance of drug in amniotic fluid, Goodman and Gilman (1955) stated that sulfadiazine appears more slowly in amniotic fluid than in fetal blood. Buniatova (1974) reported the passage of sulfapyridazine to the amniotic fluid of rat fetuses.

Prostup sulfadimidinu placentou myši a krysy

Prostup sulfadimidinu placentou myší byl sledován ve druhé polovině jejich březosti. Prostup sulfadimidinu nebyl úplný, neboť jen část množství podaného matkám prostoupila z krevního oběhu matek do fetů.

Prostup sulfadimidinu placentou krys byl sledován ve dvou stadiích březosti, a to na konci prvé její poloviny (8. až 10. dne) a ke konci březosti (18. až 20. dne). V prvém případě byl zaznamenán téměř úplný a rychlý prostup sulfadimidinu z oběhu matek do fetů. Ve druhém případě, tedy koncem březosti bylo zjištěno, že prostup sulfadimidinu placentou je výrazně omezen. Sulfadimidin byl též zjištěn v amniové tekutině.

Проницаемость сульфадимидина плацентой мыши и крысы

Проницаемость сульфадимидина плацентой мышей исследовалась во второй половине их беременности. Проницаемость сульфадимидина была неполной, ибо лишь часть вводимого количества перешла из кровообращения матерей в зародыши. Проницаемость сульфадимидина плацентой крыс исследовалась на двух стадиях беременности, а именно в конце ее первой половины (8—10 день) и в конце беременности (18—20 день). В первом случае наблюдалась почти полная и быстрая проницаемость сульфадимидина из кровообращения матерей в зародыши. Во втором случае, следовательно, в конце беременности, было установлено, что проницаемость сульфадимидина плацентой существенно ограничена. Сульфадимидин был также обнаружен в амниотической жидкости.

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