ACETYLATION OF SULFADIMIDINE AND PARA-AMINOBENZOIC ACID IN RATS DURING THE ACTION OF ANTIBIOTICS OF THE TETRACYCLINE GROUP

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The detoxication of compounds containing an aromatic amino group by acetylation is one of the most common forms of detoxication occurring in the body, especially in the liver, of man and most animals. The maintenance of the normal level of these processes is particularly important during the treatment of infections by means of drugs, when the demand for the elimination of toxic products is great and the antitoxic function of the liver, as a rule, is lowered by the infection itself.

Nevertheless insufficient attention has been paid to the study of the action of antibiotics of the tetracycline group, widely used in the treatment of infectious diseases, with these considerations in mind. The few reports in the literature in relation to chlortetracycline [2, 6] are not enough to allow comparisons to be drawn with the other members of this group of antibiotics, or to elucidate the problem of the importance of the substrate of acetylation.

We have studied the action of chlortetracycline, tetracycline, and oxytetracycline on the acetylation of para-aminobenzoic acid (PABA) and sulfadimidine in experiments on rats. The first of these substrates is a normal metabolic product of the body and the second is a compound closely related to it in chemical structure, but foreign to the animal organism.

EXPERIMENTAL METHOD

The concentration of the acetylated products of sulfadimidine and PABA was determined in the urine and blood of albino rats at various intervals after injection of the tetracyclines.

Experiments with sulfadimidine. Rats weighing 130-160 g were used in the experiments. The animals were divided into 4 groups, each of 9 rats. The rats of the first 3 groups received chlortetracycline, tetracycline, or oxytetracycline by mouth in a dose of 500 mg/kg body weight for 5 days. The animals of the fourth group did not receive antibiotics and acted as controls. On the third, fifth, and eighth days after the beginning of administration of the antibiotics, the urine of the rats was tested for its concentration of total and free sulfadimidine. For this purpose, during the 24 h before the experiment all the rats received sulfadimidine: half the animals in each group received 50 mg of the drug by mouth and the other half received 5 mg intraperitoneally, after which the animals were transferred to metabolism cages for collection of their urine. It became clear during the experiments that acetylation of sulfadimidine does not vary, even during administration of the antibiotics, so that the third period (the eighth day), chosen to determine the time taken for normal conditions to be restored, was dispensed with.

Experiments with PABA. To determine the acetylation of PABA in the urine of the animals, experiments were carried out in conditions to those of the experiments with sulfadimidine. PABA was injected intraperitoneally into all the rats in a dose of 2 mg per rat. The concentration of acetylated PABA in the urine was determined on the third, fifth, and eighth days after the beginning of administration of the antibiotics.

To study the dynamics of acetylation of these preparations in the blood stream, rats were injected with a single dose of the tetracyclines identical with that given above. Immediately after this injection, all the animals, both experimental and control (not receiving antibiotics) were given sulfadimidine or PABA, and after intervals of 1, 3, and 5 h the concentration of total and free substrate in the blood was determined.

TABLE 1. Concentration of Acetylated Sulfadimidine in the Blood of Rats After Administration of Tetracyclines (in %)

Antibiotic	Time of taking blood sample (h)					
	1		3		5	
	Mean value	Change	Mean value	Change	Mean value	Change
Chlortetracycline	8.8 ± 2.8	0.4 P > 0.05	11.9 ± 0.6	1.4 P > 0.05	10.3 ± 1.8	1.0 P > 0.05
Tetracycline	10.2 ± 4.0	1.0 P > 0.05	10.1 ± 2.4	0.4 P > 0.05	13.1 ± 3.4	1.8 P > 0.05
Ocytetracycline	9.8 ± 1.2	0.6 P > 0.05	13.6 ± 2.3	3.1 < 0.05	14.7 ± 1.8	3.4 P > 0.05
Control	9.2 ± 2.2		10.5 ± 1.9		11.3 ± 1.8	

TABLE 2. Effect of Tetracyclines on Concentration of Acetylated Sulfadimidine in the Urine of Rats (in %)

Antibiotic	Time of taking urine samples (days)			
	Third		Fifth	
	Mean value	Change	Mean value	Change
Chlortetracycline	58.9 ± 2.8	0 P > 0.05	57.1 ± 2.7	3.6 P > 0.05
Tetracycline	61.3 ± 4.4	2.4 P > 0.05	56.6 ± 2.4	4.1 P > 0.05
Oxytetracycline	59.8 ± 2.7	0.9 P > 0.05	56.2 ± 3.6	4.5 P > 0.05
Control	58.9 ± 3.5		60.7 ± 1.9	

TABLE 3. Effect of Tetracyclines on Concentration of Acetylated PABA in the Urine of Rats (in %)

Antibiotic	Time of taking urine samples (days)					
	Third		Fifth		Eighth	
	Mean value	Change	Mean value	Change	Mean value	Change
Chlortetracycline	76.1 ± 5.1	8.4 P > 0.05	65.7 ± 2.4	13.9 P < 0.05	71.6 ± 6.1	5.8 P > 0.05
Tetracycline	68.1 ± 6.4	0.4 P > 0.05	71.6 ± 4.9	8.0 P < 0.05	69.5 ± 5.8	7.9 P > 0.05
Oxytetracycline	63.0 ± 4.1	4.7 P > 0.05	75.8 ± 5.3	3.8 P > 0.05	72.3 ± 5.5	5.1 P > 0.05
Control	67.7 ± 6.8		79.6 ± 5.3		77.4 ± 4.5	

The concentrations of sulfadimidine and PABA in the test fluids were determined by the method of Timofeeva [7], which is a modification of that described by Prebsting and Gavrilov [3]. The degree of acetylation was judged by the difference between the values of the total and free compound and the acetylated produce was expressed as a percentage of the total.

EXPERIMENTAL RESULTS

Preliminary experiments were carried out in order to discover the effect of the tetracyclines on the acetylation of sulfadimidine and PABA in the blood during the first hours after administration of antibiotics, at a time when the blood concentrations of the latter were maximal. When analyzing the results of these investigations attention was directed to the low degree of acetylation of these particular substrates in the blood. For instance, in the ex-

periments with sulfadimidine, the concentration of the acetylated product determined at these times varied between 9.2 and 11.3% (Table 1). In analogous experiments with PABA, no significant difference between the concentrations of total and free PABA could be found.

The administration of a large dose of tetracyclines (500 mg/kg) to the rats had no effect on the indices of acetylation of these compounds in the blood. As is clear from the example of sulfadimidine (Table 1), the small differences in the degree of acetylation in the experimental animals were not statistically significant.

However, the low indices of acetylation of the test substrates in the blood which were obtained both in the control and in the experimental animals did not prove this point conclusively. It was necessary as the next step to determine the concentration of the acetylated products of sulfadimidine and PABA in the urine of the animals, so that the changes taking place in acetylation in the organism in the course of the 24 h period could be examined. As a result of the distribution of sulfadimidine and its conjugation as demonstrated in man, the concentration of the acetylated form in the urine is always higher than in the blood [1]. It has also been found that the urine of the rat has a high concentration of acetylated products of sulfadimidine [4] and PABA [5].

In view of these facts the main experiments to study the effect of tetracyclines on the acetylation of sulfadimidine and PABA were directed towards estimation of the concentration of these compounds in the urine of rats at various intervals after repeated administration of the antibiotics.

The results given in Table 2, showing the concentration of acetylated sulfadimidine on the third and fifth days in the urine of rats receiving tetracyclines in a dose of 500 mg/kg for 5 days, make it clear that there was no significant difference between the amount of this acetylated product in the urine of the experimental and the control animals. Special experiments also demonstrated that the degree of acetylation of sulfadimidine was independent of the mode of its administration to the animals (orally or parenterally).

It will be clear from Table 3 that on the third day of administration of the tetracyclines the process of acetylation of PABA was not significantly changed. On the fifth day (the last day of administration of the antibiotics) the concentration of acetylated PABA fell, under the influence of chlortetracycline, from 79.6% in the control animals to 65.7% ($P < 0.05$). During the same period the intensity of acetylation also showed a marked tendency to fall as a result of the action of tetracycline and oxytetracycline. These changes were still present on the eighth day, i.e., 3 days after administration of the antibiotics had ceased, although in the experiments with chlortetracycline a tendency towards normalization was observed.

These experimental results revealed a definite difference between the tetracyclines in their action on the acetylation of these two different substrates. While they had no effect on the acetylation of sulfadimidine, chlortetracycline was found to have a depressant action on the acetylation of PABA. Moreover, in the latter case, a clear difference was observed between the action of chlortetracycline and the action of the other two members of this group of antibiotics.

The fact that the acetylating function of the liver may be depressed by an antibiotic is itself interesting, for according to some workers [4, 5], this process in rats is highly resistant even to such strong agents as ionizing radiation, although these authors report that the acetylation of PABA was slightly more sensitive than the acetylation of sulfanilamide. Similar findings in respect of the tetracyclines, obtained by ourselves and also reported in the literature [2, 6] suggest that the depressant action of these antibiotics on acetylation is exhibited initially on the conversion of natural products of metabolism. Apparently the power of the liver to acetylate foreign substrates is more resistant.

SUMMARY

As a result of administration of chlortetracycline, tetracycline and oxytetracycline to rats per os in a dose of 500 mg/kg for a period of 5 days no changes were observed in the urinary content of acetylated sulfadimidine in experimental animals. In the same conditions there was a distinct reduction of the PABA acetylation under the effect of chlortetracycline, most pronounced on the fifth day (the last day of antibiotic adminstration). The data obtained pointed to a greater sensitivity of hapetic acetylating function in respect of the natural metabolite as compared to a foreign substrate.

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All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. ***Some or all of this periodical literature may well be available in English translation.*** **A complete list of the cover-to-cover English translations appears at the back of this issue.**