EFFECT OF ANTIBIOTICS OF THE TETRACYCLINE GROUP ON THE ACTIVITY OF LIVER CATALASE

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The first reports of the hepatoxic effects of tetracylines in human subjects were soon followed by experimental work designed to elucidate its mode of action. A number of papers have established the depressive effect of tetracyclines on the activity of liver enzymes [6, 10, 11, 13]. The toxic effect of these drugs on the liver may be attributed to the high capacity of the liver for binding these compounds [3, 7, 8].

We could find practically no reference in the literature to the effect of tetracyclines on the activity of enzymes participating in biological oxidation processes, and, in particular, of catalase. Apart from this, the study of this enzyme is of particular interest in connection with indications that there is a certain correlation between the functional state of the liver and the level of its catalase activity [9, 12].

The present research was devoted to a comparative study of the effects of chlortetracycline, tetracycline, and oxytetracycline on liver catalase activity, as shown by in vivo and in vitro experiments.

EXPERIMENTAL METHODS

Male rats, weighing from 130 to 160 g, were taken for the in vivo experiments. They were separated into four groups, each of 10-12 animals. Daily doses of 100 mg/kg of chlortetracycline, tetracycline, and oxytetracycline were fed to three groups, for 5 days. The fourth group served as controls, and were not given antibiotics. The rats were killed on the 5th day, two hours after the last feed, and their livers were taken for analysis.

In the in vitro experiments the hydrochlorides of the antibiotics were added to the reaction systems, at concentrations of 5, 50, and 200 μg/ml. The enzyme solution used in all the experiments was an extract of normal rat liver. We performed 4-6 experiments with each of the three antibiotics, at each of the specified concentrations.

Catalase activity was measured in both cases in the liver extracts, in the following way. A 200 mg portion of liver was ground up in a mortar with 5 ml of water, to give a homogenate. This was centrifuged, and the supernatant was taken for assay, after dilution with amounts of water recommended in the literature [2] as being optimum for the given organ. The same procedure as for blood catalase was then followed [1]. The activity was expressed as the catalase number, being the amount of hydrogen peroxide decomposed by 1 ml of solution in 30 min.

EXPERIMENTAL RESULTS

The data of Table 1 show the catalase activities of rat liver following administration of the tetracyclines at a daily dose of 100 mg/kg for 5 days.

It is evident from the data of Table 1 that chlortetracycline and tetracycline depress catalase activity to about the same extent. Oxytetracycline had practically no effect on liver catalase activity, the mean value of the catalse number being close to that found for the control group. The lowering of catalase activity following administration of chlortetracycline and tetracycline is statistically significant. The considerable scatter found in the values of the catalase numbers may to some extent be attributed to seasonal differences (the experiments were performed from March through June).

The results of the in vitro experiments in which the tetracyclines were added to the reaction systems to concentrations of 5, 50, and 200 μg/ml are given in Table 2.

The data of Table 2 show that all three tetracyclines exerted depressant effect on liver catalase activity in vitro, and that this effect became more pronounced as the antibiotic concentration was increased. It is noteworthy that although oxytetracycline showed no depressant effect in the in vivo experiments, the lowering of catalase activity was even more marked for this than for the other two antibiotics in the in vitro experiments. Thus at a concentration of 50 μg/ml only oxytetracycline gave significant lowering of catalase activity (by 22.3%), whereas at this concentration chlortetracycline and tetracycline gave no more than a tendency towards lowering of catalase activity. In these cases, the differences between the control and the experimental groups were not statistically significant. At a concentration of 200 μg/ml all three antibiotics gave statistically significant lowering of catalase activity, this effect being greatest again for oxytetracycline, and amounting to 44.8%.

TABLE 1. Catalase Activity of Rat Liver Following Oral Administration of Tetracyclines in Daily Doses of 100 mg/kg for 5 Days

Antibiotic	No. of animals	Catalase number	m	t	Percentage fall
Chlortetracycline	12	4,48 (3,11—6,22)	±0,33	2,7	19,9
Tetracycline	12	4,26 (2,35—6,51)	±0,45	2,6	23,8
Oxytetracycline	10	5,76 (4,52—6,53)	±0,25	—	No change
Control	10	5,59 (4,56—6,65)	±0,24	—	—

TABLE 2. Effect of Tetracyclines on Liver Catalase Activity in vitro

Antibiotic	Conc. of antibiotic (μg/ml)	No. of exp.	Catalase number	m	t	Percentage fall
Chlortetracycline	5	4	5,09 (4,23—5,75)	±0,33	0,8	6,1
	50	4	4,51 (4,05—5,12)	±0,29	2,5	16,8
	200	6	3,99 (3,35—4,51)	±0,23	4,3	26,4
Tetracycline	5	4	5,04 (4,17—5,66)	±0,32	0,9	6,9
	50	4	4,60 (3,96—5,13)	±0,29	2,2	15,1
	200	6	3,98 (2,94—4,56)	±0,25	4,1	26,6
Oxytetracycline	5	4	5,02 (4,10—5,59)	±0,33	1,0	7,4
	50	4	4,21 (3,59—4,71)	±0,23	3,7	22,3
	200	6	2,99 (2,19—3,59)	±0,21	7,6	44,8
Control	—	6	5,42 (4,47—6,02)	±0,24	—	—

It should be noted that some inactivation of the enzyme may have been due, in the in vitro experiments, to the lowering of the pH of the medium as a result of adding the tetracycline derivatives. In order to clear up this point we measured the pH of the systems. We found that the pH fell from 6.5 before adding antibiotics to a concentration of 200 μg/ml to 3.6 after the addition. Measurement of catalase activity in media not containing antibiotics showed that it fell by not more than 12% when the pH was lowered from 6.5 to 3.6.

For the interpretation of the results of the in vivo and in vitro experiments it is necessary to take into account the concentrations of the antibiotics in the liver of animals to which they had been administered. According to G. Ya. Kivman [3] and to G. Ya. Kivman and N. M. Smol'nikova [5], oxytetracycline is bound less firmly by liver tissue than are the other two antibiotics. Experiments with homogenates showed that liver tissue binds only half the amount

of oxytetracycline than it does of chlortetracycline or of tetracycline. Another factor is that oxytetracycline is absorbed from the gastrointestinal tract to a lesser extent following its oral administration than are the other two antibiotics [4].

Because of these factors, the concentration achieved in the liver would be smaller in the case of oxytetracycline than in those of the other two antibiotics, when they are fed at equal dosage levels; the effective concentration may, evidently, be so low as not to give any appreciable effect on catalase activity.

SUMMARY

Tetracyclines were given to rats per os in a dose of 100 mg/kg for a period of 5 days. Chlortetracycline and tetracycline caused a reduction of hepatic catalase activity. Oxytetracycline had practically no effect on this enzyme. In experiments with the addition of antibiotics into the reagents' mixture in concentrations of 5, 50 and 200 μg/ml all the 3 tetracyclines decreased the activity of the hepatic catalase, this action being the most pronounced with oxytetracycline. No action of the latter obtained in vivo may be attributed to its lesser absorption faculty in the gastrointestinal tract and binding capacity to the hepatic tissue.

LITERATURE CITED

1. A. N. Bakh and S. R. Zubkova, Collected Works of Academician A. N. Bakh [in Russian] (Leningrad, 1937), p. 411.
2. G. V. Voskoboinikov, Med. Radiol. No. 1 (1956), p. 39.
3. G. Ya. Kivman, Antibiotiki, No. 3 (1956), p. 42.
4. G. Ya. Kivman and N. M. Smol'nikova, Antibiotiki, Vol. 6, No. 8 (1961), p. 702.
5. G. Ya. Kivman and N. M. Smol'nikova, Byull. Éksper. Biol. i Med. No. 12 (1959), p. 68.
6. V. Ya. Kudryavtseva, Antibitotiki, No. 2 (1960), p. 58.
7. L. E. Böttiger, Acta. Med. Scand. Vol. 151 (1955), p. 343.
8. S. Helander and L. E. Böttiger, Ibid. Vol. 147 (1953), p. 71.
9. S. Iagnov, Acta. Med. Acad. Sci. Hung. Vol. 10, No. 3 (1957), p. 183.
10. S. N. Iyer, K. L. Arora, and C. R. Krishna-Murti, J. Scient. and Industr. Res. B., Vol. 12B (1953), p. 536.
11. W. F. Loomis, Science, Vol. 111 (1950), p. 474.
12. M. Takahashi, Tohoku Igakuzassi, Vol. 58 (1958), p. 54.
13. Hg. Zimmerman and F. Humoller, Am. J. Physiol. Vol. 175 (1953), p. 468.

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