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Effects of ethyl chlorophenoxyisobutyrate (CPIB) on hepatic catalase activity of mice bearing Ehrlich ascites tumor cells*

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SUMMARY

The catalase activity of livers in mice bearing Ehrlich ascites tumor cells was maintained at a normal level by the oral administration of CPIB throughout the tumor-bearing course. From the results of injection experiments of CPIB, it was noted that the rate of increase in catalase activity varied considerably at different stages of the tumor-bearing course.

INTRODUCTION

The marked diminution in catalase activity of livers of tumor-bearing animals was established by Greentein[3]. Although there are extensive investigations of this enzyme since then, the precise mechanism of the reduction of the activity is still obscure. Recent immunochemical researches have, however, shown that this phenomenon was due to the absolute diminution in catalase protein, suggesting a depressed rate of biosynthesis of the enzyme[5,9].

On the other hand, a hypolipidemic agent, ethyl-α-*p*-chlorophenoxyisobutyrate (CPIB), has become known to be capable of enhancing catalase activity as well as proliferating peroxisomes[4,13], probably the result of an increased biosynthesis of catalase[1,11]. Thus it becomes of interest to investigate whether animals bearing tumors would respond to CPIB. The present studies report the effect of CPIB on catalase activity in the liver of mice bearing Ehrlich ascites tumor cells.

MATERIAL AND METHOD

Male JCL/ICR mice weighing between 25 and 30 g were used in these

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experiments. Ehrlich ascites tumor cells of a hyperdiploid line[15] were inoculated intraperitoneally into mice at a dose of 1×10^7 cells[2,8,14]. In the feeding experiments, mice were maintained on a diet containing 0.25% CPIB from the time of tumor inoculation to the time of exangination. Tumor-bearing mice fed on a normal diet and mice treated with CPIB alone were used as controls. They were sacrificed by decapitation on the 3rd, 5th, 7th, 10th and 14th day of treatment. In the injection experiments, CPIB was diluted with olive oil to provide 500 mg/ml and injected subcutaneously into mice at a dose of 25 mg CPIB once a day for four consecutive days. The injections were started on the 3rd, 5th 7th and 10th day after the inoculation of tumor cells. The control group was injected in the manner described above without inoculation with tumor cells. After decapitation, the livers were quickly removed and homogenized in ice cold distilled water. Catalase activity was measured by the titrimetric method of Adams with a slight modification as previously described[16], and values were expressed on the the basis of units per 100 mg of dry tissue.

RESULTS

The results are shown in Figs. 1 and 2.

Tumor-bearing mice: Catalase activity in livers of this group showed a gradual decrease as the tumor-bearing course advanced, and reached a critical point on the 7th day after the inoculation. Subsequently, the activity was maintained at about half values of normal liver.

Mice fed on CPIB without tumor: Catalase activity of livers of mice fed on 0.25% CPIB for 3 days increased by about 20%, and further increases in the activity were observed till the 7th day of CPIB treatment. Thereafter, no further increse was seen despite the continuation of CPIB-feeding.

Tumor-bearing mice fed on CPIB: Catalase activity neither incresed nor decreased after treatment for 3 days to tumor-bearing mice. The enzyme activity appeared to be slightly increased after 5 days of treatment. After 7 days and 10 days of treatment, catalase activity showed the lowest level during the treatment, and after 14 days of administration of CPIB a slight increase in the activity was observed, although the animals were cachectic. Catalase activity in livers of tumor-bearing and CPIB-treated mice appeared to be somewhat changeable, but the difference were not significant at a 5% level.

Mice injected with CPIB alone: Catalase activity of liver of mice

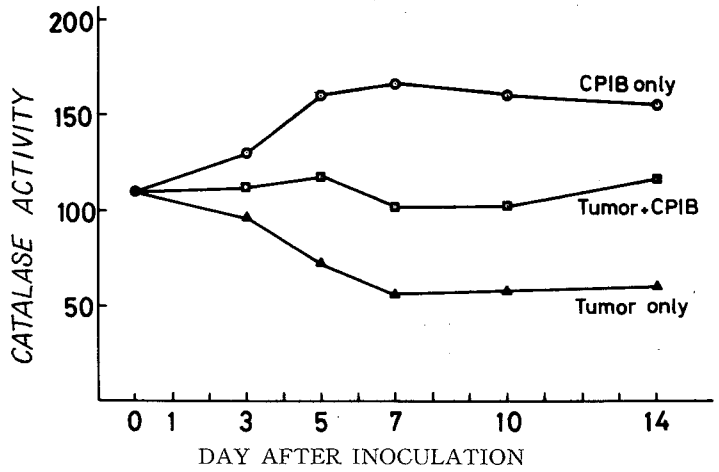

Fig. 1. Effect of CPIB feeding on hepatic catalase activity of mice with or without Ehrlich ascites tumor. Black circle; normal liver. Open circles; livers of mice fed on CPIB. Quadrangles; livers of mice treated with both tumor and CPIB. Triangles; livers of mice bearing tumor. Each point consisted of five or more mice.

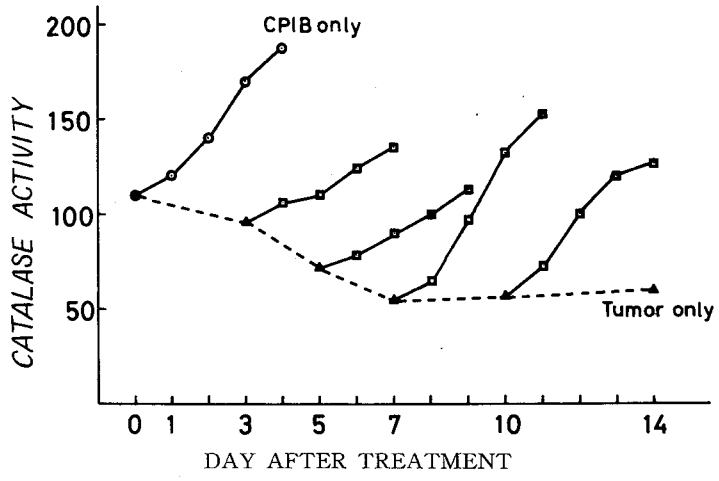

Fig. 2. Effect of CPIB injection on hepatic catalase activity of mice with or without Ehrlich acites tumor. Black circle; normal liver. Open circles; livers of mice injected with CPIB. Quadrangles; livers of mice treated with both tumor and CPIB. Triangles; livers of mice bearing tumor. Each point consisted of three or more mice.

without tumor increased in accordance with injections of CPIB. After four injections, it reached 170% of normal values. The average increase rate was calculated to be about 20 units per day.

Tumor-bearing mice injected with CPIB: When the injection was started on the 3rd day after tumor inoculation, the activity increased gradually and the rate of increase in the activity was about half of that of normal. When the injection was started on the 5th day, the activity increased at a rate similar to that started on the 3rd day after tumor inoculation. When started on the 7th day, the increase was very rapid. The rate of increase slightly exceeded that in control. When started on the 10th day, the rate of increase was similar to the rate in control.

DISCUSSION

The catalase activity in the host liver was prevented from decreasing by oral administration of CPIB throughout the tumor-bearing course, as seen in Fig. 1. Moreover, as was evident from Fig. 2, the decreased catalase activity by the presence of tumors was markedly increased by the injection of CPIB.

The depressed catalase activity in livers of tumor-bearing animals has been considered to be due to the decreased rate of its biosynthesis, not only from *in vivo* experiments[5,9,10], but also from *in vitro* experiments[6,7,17,18]. However, the results presented here suggest strongly that the depressed rate of catalase biosynthesis may be reversible.

During the present investigations, Reddy *et al*[12] reported the effect of CPIB on hepatic catalase of rat bearing subcutaneous tumors, but their examination was conducted after a prolonged administration of CPIB. In the present study, emphasis was laid on the effect of CPIB at different stages of tumor-bearing course. It is clearly shown from Fig. 2 that the rate of increase in catalase activity showed a considerable variation at different stages of the tumor-bearing course. The increase rate of catalase activity in the mice injected from the 3rd and the 5th day of tumor inoculation, was about half of the control, while that in mice injected from the 7th day slightly exceeded that of control. As already reported[2,8,14], during the first week after the inoculation, Ehrlich ascites tumor cells proliferated exponentially in the peritoneal cavity, and the catalase activity decreased progressively. Thereafter, the growth of tumor cells showed a stationary phase and the catalase activity showed a new lowered steady state. Thus, it is of particular interest that the rate of increase in catalase activity by CPIB was slow during the exponential growth of tumor cells, and that the activity increased at a similar

rate to the control during the steady state proliferation of tumor cells.

In the present investigations, the mechanism of the depression of liver catalase activity in tumor-bearing animals could not be clarified. However, it is important to note that, even at the terminal stage of tumor development, in which mice were cachectic, the catalase which are regarded as non-essential enzyme was induced at a similar rate to the control mice. A considerable amount of time and effort must be spent to clarify the mechanism by which the catalase activity is depressed in livers of tumor-bearing animals.

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