FACTORS IN POSTTRANSCRIPTION REGULATION OF THE RATE OF LIVER CATALASE SYNTHESIS IN RATS WITH TUMORS

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The state of factors controlling the rate of catalase synthesis in the rat liver at posttranscription stages under normal conditions and during the development of a malignant tumor (2nd, 7th, and 14th days after inoculation of Pliss lymphosarcoma) was investigated. A simple method is suggested for determining the biological activity of two factors – activator (F_{act}) and inhibitor (F_{in}). The results showed that if $F_{act}/F_{in} \geqslant 1$, catalase synthesis is observed (2nd and 14th days after inoculation of the tumor), but if $F_{act}/F_{in} < 1$, synthesis of the enzyme is completely suppressed (7th day after inoculation of the tumor).

KEY WORDS: *catalase; regulation of synthesis; carcinogenesis.*

The total catalase activity of the liver in animals with tumors is known to be much lower than normal [6, 7], and according to some investigators this is the result of the effect of the tumor on enzyme synthesis [2, 9]. A study of the mechanism of inhibition of catalase synthesis in Morris hepatoma 5123 C showed that this process is regulated at the posttranscription stages by two cytoplasmic factors, one of which stimulates catalase synthesis (F_{act}), whereas the other inhibits it (F_{in}) [12-15].

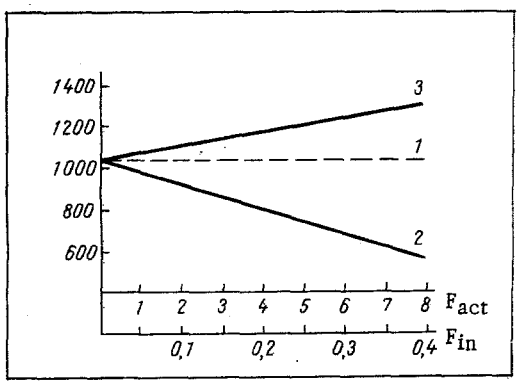

Fig. 1. Determination of activity of F_{act} and F_{in}: 1) control (increase in catalase activity 48 h after injection of AT); 2) increase in catalase activity after injection of F_{in}; 3) increase after injection of F_{act}. Abscissa, concentrations of F_{act} and F_{in} in mg protein; ordinate, catalase activity (in μmoles substrate in 1 ml cytosol fraction in 1 min).

The object of this investigation was to study the state of the factors of posttranscription regulation of the rate of catalase synthesis in the liver of rats inoculated with Pliss lymphosarcoma.

EXPERIMENTAL METHOD

Noninbred male rats weighing 160-180 g were used. Cytoplasmic factors were isolated by the method of Uenoyama and Ono [14,15] and analyzed for their protein content by Lowry's method [8], for their content of SH groups by amperometric titration [4], and for their catalase activity by a manometric method [1]. Tests were carried out on intact animals and also on the 2nd, 7th, and 14th days after subcutaneous inoculation of Pliss lymphosarcoma. To calculate the activity of the two factors, i.e., their ability to stimulate or inhibit catalase synthesis, the following method was used. Intraperitoneal injection of 3-amino-1,2,4-triazole (AT) led to blocking of all tissue

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TABLE 1. Activity of Factors Controlling Rate of Catalase Synthesis in Rat Liver (μmole/min/g liver) under Normal Conditions and during Carcinogenesis

Time after inoculation of tumor (in days)	F_{act}	F_{in}	F_{act}/F_{in}
Control	625	200	3,14
2	1250	702	1.78
7	465	825	0 57
14	1147	720	1,59

catalases, and any further increase in the activity of this enzyme with time could only have been due to its synthesis *de novo* [2, 10, 11]. Knowing the catalase activity in time, the effect of the factors on catalase synthesis could be determined if injected intravenously after a preliminary injection of AT. AT was accordingly injected intraperitoneally (1 g/kg body weight), and 24 h later 0.2, 0.6, and 1.2 ml of F_{act} and F_{in}, obtained by chromatography on DEAE-cellulose, was injected into the caudal vein of the rats. When 24 h had elapsed after the injection of the factors, catalase activity was calculated graphically on the basis of the concentrations of preparations injected and the corresponding levels of catalase activity. The tangent of the angle of slope of the resulting straight lines was the measure of catalase activity in unit time in response to the action of 1 mg F_{in} or F_{act} (Fig. 1).

EXPERIMENTAL RESULTS AND DISCUSSION

Data showing the state of catalase synthesis in the liver of intact rats and rats with tumors are given in Tables 1 and 2. They show that the substances isolated were biologically active preparations capable of stimulating or inhibiting catalase synthesis *in vivo*. In hepatoma [12-15] activity of F_{in} is increased and activity of F_{act} is reduced. As Table 1 shows, a sharp increase in F_{in} activity in the liver of a tumor-bearing animal also was observed on the second day after inoculation of the tumor, i.e., the effect of the malignant neoplasm on F_{in} was not dependent on its localization. As regards F_{act}, its activity in the liver at this time was greater than normal. As a result of dissimilar change in the activities of the factors (the rate of change of F_{in} was much greater), the ratio F_{act}/F_{in} fell below the normal level but still remained higher than unity. As growth of the tumor progressed, by the 7th day its influence on the host tissues grew stronger: F_{act} fell but, at the same time, the value of F_{in} increased still further, so that the F_{act}/Fin ratio fell to a minimum. By the 14th day the ratio showed some increase, although it still remained lower than normal. It was shown previously that catalase synthesis in the liver is reduced to one-tenth of its initial value on the 2nd day after inoculation of lymphosarcoma, falls to zero on the 7th day, and rises a little on the 14th day [3]. The present investigation showed correlation between inhibition of catalase synthesis in the liver and the F_{act}/Fin ratio at the same time after transplantation of the tumor. It will be clear from Table 1 that the ratio between the activities of these factors fell by the 2nd day, reached a minimum on the 7th day, and then increased a little by the 14th day. If the F_{act}/F_{in} ratio were equal to or greater than unity, some catalase at least must be synthesized in the liver. If, however, its value fell below unity, synthesis of the enzyme would be completely suppressed. As a result of activation of F_{in}, the latter may become associated with catalase-synthesizing ribosomes, and could consequently cause the inhibition of protein synthesis [13-15].

To study the possible mechanism of action of the regulators, their content of protein and SH groups was calculated (Table 2). It follows from Tables 1 and 2 that activation of

TABLE 2. Content of Protein and SH Groups in Factors Controlling Rate of Catalase Synthesis in Liver of Rats under Normal Conditions and during Carcinogenesis

Time after inoculation of tumor (in days)	SH groups (in moles/mole protein)		Protein concentration (in mg/ml)	
	F_{act}	F_{in}	F_{act}	F_{in}
Control	38,4±2,08	19,2±1,92	0,123±0,015	0,073±0,007
7	28,2±4,4	27,0±3,02	0,115±0,005	0,183±0,020
14	37,2±3,2	41,2±2,96	0,125±0,011	0,112±0,009

Legend. When content of SH groups per mole protein was calculated, molecular weights of factors were taken from [14] (M±m)

F_{in} during carcinogenesis correlated with the content of protein and SH groups. This result is in agreement with the observations of Spirin [5] who showed that the ability of proteins to associate with ribosomes correlates with the content of SH groups in the proteins. As regards F_{act}, no correlation was found between its activity and the content of protein and SH groups in carcinogenesis, but it must be pointed out that the hypothetical mechanism of action of this factor may be in the reassociation of F_{in} with ribosomes [14, 15].

The results described above not only explain the fact that a given malignant neoplasm can influence the liver catalase and enable that influence to be assessed qualitatively, but they also enable the factors controlling catalase synthesis and the dynamics of their changes during tumor growth to be expressed quantitatively.

LITERATURE CITED

1. A. N. Bakh, Selected Works [in Russian], Moscow (1937), p. 12.
2. Z. Hruban and M. Rechcigl, Microbodies and Related Structures, Academic Press, New York (1969).
3. V. P. Komov and T. F. Rakhmanina, Vopr. Onkol., No. 11, 48 (1974).
4. V. V. Sokolovskii, Lab. Delo, No. 8, 3 (1962).
5. A. S. Spirin, Uspekhi Sovr. Biol., 77, No. 2, 155 (1974).
6. S. Fujimoto, Cancer (Philadelphia), 19, 844 (1966).
7. J. P. Greenstein, J. Nat. Cancer Inst., 15, 1603 (1955).
8. O. H. Lowry et al., J. Biol. Chem., 193, 265 (1951).
9. E. T. Nishimura, T. Y. Kabara, J. P. Kaltenlach, et al., Arch. Biochem., 97, 589 (1962).
10. V. Price, W. Sterling, W. Tarantola, et al., J. Biol. Chem., 237, 3468 (1962).
11. M. Rechcigl and V. Price, Prog. Exp. Tumor Res. (Basel), 10, 112 (1968).
12. K. Uenoyama and T. Ono, Internat. J. Cancer, 9, 608 (1972).
13. K. Uenoyama and T. Ono, J. Molec. Biol., 65, 75 (1972).
14. K. Uenoyama and T. Ono, J. Molec. Biol., 74, 439 (1973).
15. K. Uenoyama and T. Ono, J. Molec. Biol., 74, 453 (1973).