ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN—POLONIA
VOL. XXV, 1 SECTIO C 1970

Z Katedry Biochemii Wydziału Biologii i Nauk o Ziemi UMCS
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The Effect of Melanoma Tumor on Catalase Activity in Hamsters

Wpływ tumoru melanoma na poziom katalazy w wątrobie u chomika

Влияние тумора *melanoma* на уровень каталазы в печени хомяка

INTRODUCTION

In 1910, Blumenthal and Brahn (1) observed a decreased liver catalase activity in tumor-bearing animals. The effect can be induced by spontaneous and transplanted tumors of various types. Most of previous experiments were carried out on mice and rats.

In this work, variations of liver catalase activity in Syrian hamsters in relation to the time of growth and weight of transplanted melanoma tumor were determined.

MATERIAL AND METHODS

The tissue of melanoma tumor, which developed spontaneously in a hamster of laboratory breeding in the Department of Biology, Medical Academy, Gdańsk, in 1959 and was described by Bomirski et al. (2), served as experimental material. For transplantation of melanoma hamsters were injected subcutaneously with 1 ml of tumor tissue homogenate in physiological saline solution. In 1 ml of the homogenate 300 mg of fresh tumor tissue without any necrotic changes was contained.

Determination of Catalase Activity. The activity of catalase both in the liver and in the tumor tissue was determined by Euler-Josephson's (3) method. Livers were taken from killed and exsanguinated animals and washed through the *vena porta* with cooled physiological saline solution buffered to *pH* 6.8. The washed liver was homogenized in physiological saline solution at *pH* 6.8 at the ratio of 65 mg of fresh tissue per 1 ml of solution in a Waring Blendor for 7 min. at 12500 r.p.m. at 0°C. The homogenate was then centrifuged for 15 min. at 10000 × g

in cold. In the supernatant catalase activity was determined towards 0.01 n H_2O_2 at *pH* 6,8. Thus, 1 ml of suitable diluted supernatant from the liver homogenate at 0°C was added to 50 ml of the substrate solution. After 3, 6, 9 and 12 min., 5 ml of this mixture was taken and catalase activity was inhibited by introducing 5 ml of 2 n H_2SO_4. The concentration of non-decomposed H_2O_2 in the sample was determined by titration with 0.005 n $KMnO_4$. Next the value of reaction constant k was calculated from the following equation

$$k = \frac{1}{t} \log \frac{a}{a - x} \text{ where}$$

k — reaction constant,
t — time in minutes,
a — substrate concentration at zero time (in ml of 0.005 n $KMnO_4$),
$(a—x)$ — substrate concentration after the time t (in ml of 0.005 n $KMnO_4$).

The k values obtained for different reaction times were graphically interpolated for the reaction time $t = 0$ and conventional catalase activity was expressed in generally accepted units:

$$\text{Kat. f} = \frac{k_o \, x \text{ enzyme dilution}}{\text{protein in g/ml}}$$

Protein was determined in parallel assays by Lowry's method (5). The determination of catalase in the tumor tissue was carried out in the same way as in the liver.

RESULTS

Liver catalase activity in melanoma-bearing hamsters was determined every 7 days during a 49-day-period, which was the average survival time of the tumor-bearing animals. Simultaneously, determinations were made on healthy hamsters as controls. Values of the decrease of liver catalase activity in relation to the time of tumor growth are shown in Fig. 1, whereas those of liver catalase activity in relation to the weight of the tumor tissue are illustrated in Fig. 2. The values of the so-called catalase depression were calculated after Riley (6):

$$\text{depression } \% = \frac{\Sigma_1 - \Sigma_2}{\Sigma_2} \cdot 100$$

where

Σ_1 — mean liver catalase activity of controls
Σ_2 — mean liver catalase activity in animals bearing melanoma tumors

The liver catalase depression in relation to the time of tumor growth increased slowly (Fig. 3), whereas the values of depression calculated in relation to tumor tissue weight increased more progressively (Fig. 4).

The catalase activity in the tumor tissue is about a hundred times lower than in the liver tissue and it increases slowly with the time of tumor growth (Table 1).

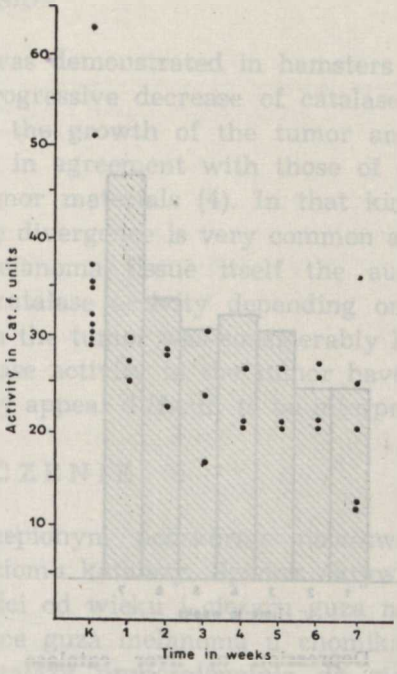

Fig. 1. The level of liver catalase activtiy in hamsters with transplanted melanoma in relation to the time of the tumor growth. K = control

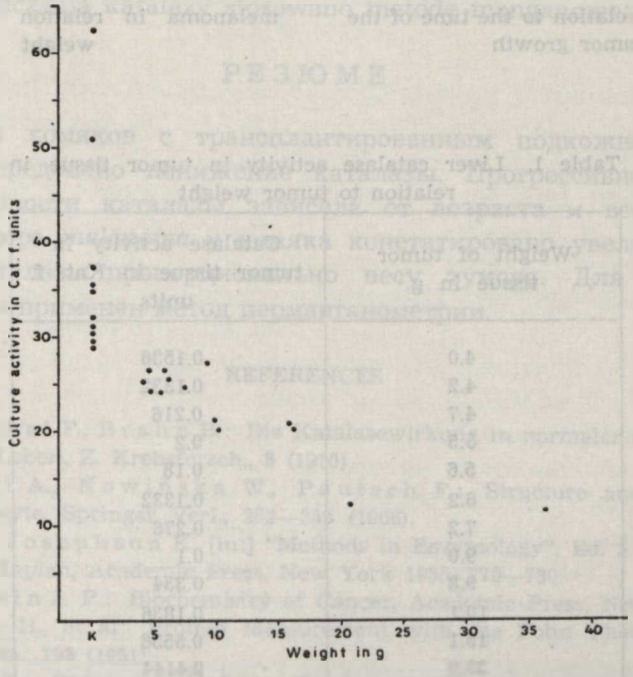

Fig. 2. The level of liver catalase activity in hamsters with transplanted melanoma in relation to the weight of the tumor tissue. K = control

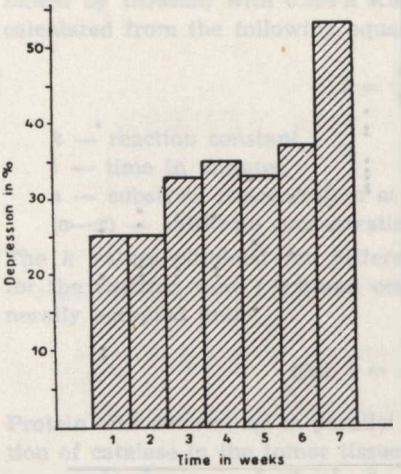

Fig. 3. Depression of liver catalase activity in hamsters with transplanted melanoma in relation to the time of the tumor growth

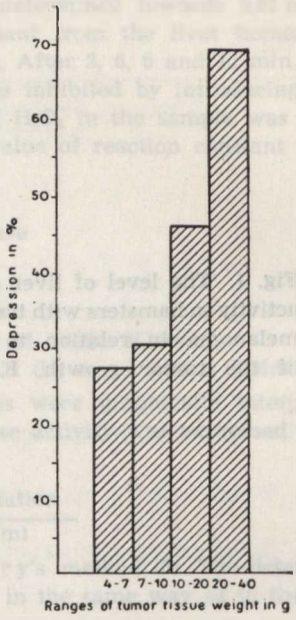

Fig. 4. Depression of liver catalase activity in hamsters with transplanted melanoma in relation to the tumor weight

Table 1. Liver catalase activity in tumor tissue in relation to tumor weight

Weight of tumor tissue in g	Catalase activity in tumor tissue in Kat. f. units
4.0	0.1536
4.2	0.1332
4.7	0.216
5.5	0.2
5.6	0.18
6.2	0.1332
7.2	0.276
9.0	0.1
9.8	0.334
10.1	0.1836
15.1	0.5556
23.0	0.4144
36.0	0.4592

DISCUSSION

Liver catalase activity decrease was demonstrated in hamsters with transplanted melanoma tumor. A progressive decrease of catalase was observed in relation to the time of the growth of the tumor and its weight increase. These results are in agreement with those of other authors on different animal and tumor materials (4). In that kind of studies the occurrence of considerable divergence is very common as far as individuals are concerned. In melanoma tissue itself the authors observed a progressive increase of catalase activity depending on the tumor weight. The level of catalase in the tumor was considerably lower than in the liver. Variations of catalase activity in the tumor have not been described yet and at present they appear difficult to be interpreted.

STRESZCZENIE

W wątrobie chomików z przeszczepionym podskórnie nowotworem „melanoma" stwierdzono obniżkę poziomu katalazy. Spadek aktywności katalazy był progresywny w zależności od wieku i ciężaru guza nowotworowego. Natomiast w samej tkance guza melanoma u chomika zaobserwowano wzrost aktywności katalazy proporcjonalnie do ciężaru guza. Do oznaczania katalazy stosowano metodę manganometryczną.

РЕЗЮМЕ

В печени хомяков с трансплантированным подкожно тумором *melanoma* определено понижение каталазы. Прогрессивность понижения активности каталазы зависела от возраста и веса тумора. В ткани тумора *melanoma* у хомяка констатировано увеличение активности каталазы пропорционально весу тумора. Для определения каталазы применен метод перманганометрии.

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Papier druk. sat. III kl. 80 g Format 70 × 100 Druku str. 6
Annales UMCS Lublin 1969 Lub. Zakł. Graf. Lublin, Unicka Zam. 820. 18.III.70
1100 + 25 egz. Z-6 Manuskrypt otrzymano 18.III.70 Data ukończenia 30.XII.70