Inhibition of the Effect of Some Carcinogens by Their Partially Hydrogenated Derivatives

It has been observed in various fields of biochemistry that a biologically active compound—for example, a vitamin—may be prevented from displaying its effect in the presence of a closely related derivative (antivitamin). The latter probably plays the part of a competitor for intracellular receptors.

The basic idea of our work (*1*) was to investigate whether or not the simultaneous injection of a strong carcinogen and a close derivative that differs from the carinogen by the level of oxidation only would prevent the appearance of malignant tumors. Evidently these experiments had to be conducted under conditions that secured a high tumor incidence in the absence of the added compound.

For this purpose, some reduction products of 20-methylcholanthrene and of 1,2,5,6-dibenzanthracene were prepared by partial and by total catalytic hydrogenation (*2*) and subsequent chromatographic resolution. Thus, methylcholanthrene yielded the 6,7-dihydro and the 1,2,3,4,11,14-hexahydro derivatives that were described earlier by Fieser and Hershberg (*3*), as well as the fully hydrogenated perhydromethylcholanthrene. Starting from dibenzanthracene, a dihydro, a decahydro, and the perhydro derivatives were obtained; however, the structure of the two partially hydrogenated compounds cannot be established with certainty at the present time. The decahydro compound melts sharply at 176°C, while the dihydro derivative is possibly a mixture of isomers.

In each instance, 30 male C57 mice, 3 to 4 months old were treated (*4*). They were kept on a standard commercial chow diet supplemented with rolled barley and were given water *ad libitum*.

The subcutaneously injected ethyl laurate solution (single injection) contained 30 μg (1 part) of carcinogen (TD_{75}) and 15 parts of hydrogenated carcinogen. The experiments were allowed to continue for 12 months.

The results are summarized in Table 1; all tumors listed there were subcutaneous spindle cell sarcomas. Table 1 demonstrates that while the addition of the fully hydrogenated substances had no influence on the carcinogenic potency, some partially reduced derivatives were highly effective in decreasing the tumor incidence. Thus, in the methylcholanthrene series, the tumor yield was decreased from 28 percent to 8 percent. In the case of dibenzanthracene, the results were more striking since its partially hydrogenated derivatives completely inhibited the tumorigenic power of the parent compound.

The incidence data reported are in good accordance with some other features. Indeed, an association may be observed between the tumor yield, tumor induction period, intervals of subsequent tumor formation, and the chemical nature of the hydrogenates. Table 1 shows that the presence of hexahydromethylcholanthrene not only decreased the number of observed tumors from 8 to 2 but also lengthened the induction period from 21 to 31.5 weeks. Furthermore, the interval between the first and the second tumor was extended from 0 to 18 weeks.

Although the available data are not extensive enough for a broad generalization, it is clear that in the series of polycyclic carcinogens a strong inhibiting effect of partially reduced derivatives does exist and that it reaches its optimum at a certain hydrogenation level. The weak effect of dihydromethylcholanthrene may be understood on the basis of its slight structural difference from methylcholanthrene and hence by its possible *in vivo* conversion into the carcinogen. In contrast, the inertness of the two fully hydrogenated substances studied could well be explained by the circumstance that since they are void of aromatic character and are no longer close derivatives of the parent compound, they cannot act as anticarcinogens in the tissue.

Our results are in line with some related data, especially with the concept of competition between carcinogens and anticarcinogens for available intracellular receptors (*5*).

Table 1. Inhibition and retardation of the carcinogenic effect of 20-methylcholanthrene and of 1,2,5,6-dibenzanthracene by some hydrogenated derivatives.

Compound	No. of animals	Survivors at time of first tumor	No. of tumors	Sequence of tumor appearance (wk)
20-Methylcholanthrene (MC)	30	29	8	21, 21, 21, 22, 22.5, 27.5, 28.5, 34.5
6,7-Dihydromethylcholanthrene + MC	30	29	6	18, 18, 18.5, 19, 26, 50.5
1,2,3,4,11,14-Hexahydromethylcholanthrene + MC	30	30	2	31.5, 49.5
Perhydromethylcholanthrene + MC	30	30	7	14.5, 18, 27.5, 27.5, 28, 31.5, 33
1,2,5,6-Dibenzanthracene (DB)	30	29	7	19, 25.5, 27, 29, 32, 39, 43
Dihydrobenzanthracene + DB	30	30	0	none
Decahydrobenzanthracene + DB	30	29	0	none
Perhydrobenzanthracene + DB	30	30	5	27, 34, 40, 42, 44

PAUL KOTIN
HANS L. FALK
Departments of Pathology and *Biochemistry, University of Southern California School of Medicine*, and *the Laboratories, Los Angeles County General Hospital*

W. LIJINSKY
L. ZECHMEISTER
Gates and Crellin Laboratories of Chemistry, California Institute of Technology, Pasadena

References and Notes

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2. For some experimental conditions, see W. Lijinsky and L. Zechmeister, *J. Am. Chem. Soc.* 75, 5495 (1953).
3. L. F. Fieser and E. B. Hershberg, *ibid.* 60, 940 (1938).
4. The mice were obtained from the Cancer Research Genetics Laboratory, Berkeley, Calif.
5. H. G. Crabtree, *J. Pathol. Bacteriol.* 51, 303 (1940); A. Lacassagne, Buü-Hoi and G. Rudali, *Brit. J. Exptl. Pathol.* 26, 5 (1945); W. T. Hill *et al., Cancer Research* 11, 892 (1951); P. E. Steiner and H. L. Falk, *ibid.* 11, 56 (1951).

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Two Methods of Obtaining Least Squares Lines

S. I. Askovitz reported a method for determining the mean y value ($\bar{y}$) from graphic data (*1*). Two methods of obtaining the best fitting straight line through a set of points are presented here. The first method uses the Askovitz technique to find $\bar{y}$, while the slope is calculated from the values of y on a transposed x-axis. The second method is, to my knowledge, original and is completely graphic. In it, two points of the best fitting line are found directly on the graph. In each case, if the original data are in the form of a continuous curve, discrete values of x must be chosen, and the same limitation holds as in Askovitz' method, namely, that the individual points must be at equal intervals of x.

First method. A modification of the method of Arkin and Colton (*2*) is used —that is, when there are an odd number