only by a direct conversion of the fatty acid carbon.

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ENZYMES OF THE CITRIC ACID CYCLE IN TUMORS[1]

Sir:

As part of an investigation of the occurrence of the citric acid cycle in tumors, assays were made of various enzymes concerned in this process. The present report demonstrates the presence, in three transplanted mouse tumors, of the three enzymes directly involved in the metabolism of the tricarboxylic acids, *viz.*, "condensing enzyme," aconitase and isocitric dehydrogenase. The "condensing enzyme" is of particular interest because it is responsible for the initial reaction of the cycle, the formation of citrate by condensation of "active acetate" with oxalacetate.[2,3]

The data in the table indicate that the enzyme is present in the three tumors in amounts comparable with normal tissue. The other two enzymes were present also in significant amounts. Extension of these studies to other tumors and other enzymes of the cycle is under way.

TABLE I

ASSAY OF TUMORS FOR ENZYMES

	Condensing enzyme[a] μM. Citrate per 10 minutes per 100 mg. acetone powder	Aconitase[b] Units per mg. dry wt. of tissue	Isocitric dehydrogenase[c] Units per mg. acetone powder
Mouse liver	1.53[d]	33	10.8
Rhabdomyosarcoma	1.45	2.2–5.0[e]	6.4
Hepatoma	2.90	8.3	14.8
Mammary adenocarcinoma	3.30	3.1–5.9[e]	16.0

[a] For condensing enzyme the solution contained: 1 *M* KH_2PO_4 buffer, *p*H 7, 0.025 ml., 0.08 *M* $MgCl_2$, 0.05 ml., 0.2 *M* cysteine (neutralized) 0.05 ml., 0.14 *M* oxalacetic acid, 0.05 ml., 0.1 *M* synthetic acetyl phosphate, 0.10 ml., extract of *E. Coli* 4157 (from washed, lyophilized cells) 0.20 ml. To this was added 0.50 ml. of a 1–5 extract of an acetone powder of the tissue, and after incubation for ten minutes at 40°, the suspension was deproteinized and citrate determined by the method of Natelson, *et al.*[4] [b] Aconitase was determined by an unpublished method of Racker using citrate as substrate and measuring formation of *cis*-aconitate by increase in absorption of light at 240 mμ, using a Beckman spectrophotometer. The assay was made in a quartz absorption cell containing phosphate buffer, *p*H 7.4, 0.05 *M*, citrate, 0.03 *M*, and tissue extract in a total volume of 3 ml. The extract was prepared by homogenizing the tissue with 20 volumes of 0.1 *M* phosphate buffer, *p*H 7.4, and centrifuging off the residue. A unit of enzyme activity is the amount producing a change in optical density at 240 mμ of 0.001 per minute at 25°. [c] Isocitric dehydrogenase was determined on extracts of acetone powders by the optical method of Mehler, *et al.*,[5] a unit of activity being defined as the amount required to produce a change in optical density of 0.01 per minute at 25°. [d] Citrate formation in complete system without tissue was 0.21 μM. Citrate content of all tissues was <0.05 μM. [e] Three determinations on different tumors.

It might be assumed from these results that the failure of oxalacetate to be oxidized by tumor homogenates, observed by Potter and LePage,[6] may be due to loss of some necessary factor in homogenization rather than to an inability of the intact tissue to oxidize oxalacetate. We emphasize, however, that the data reported apply only to the tumors studied and generalizations are as yet unwarranted.

(1) Aided by grants from the American Cancer Society, recommended by the Committee on Growth; and the National Cancer Institute, U. S. Public Health Service. The aid of Drs. Stern and Ochoa in supplying details of the condensing enzyme assay and providing the *E. coli* and acetyl phosphate is acknowledged with deep appreciation.

(2) Stern and Ochoa, *Fed. Proceedings*, **9**, 234 (1950).

(3) Chou, Novelli, Stadtman and Lipmann, *ibid.*, **9**, 160 (1950).

(4) Natelson, Pinous and Lugovoy, *J. Biol. Chem.*, **175**, 845 (1948).

(5) Mehler, Kornberg, Grisolia and Ochoa, *ibid.*, **174**, 961 (1948).

(6) Potter and LePage, *ibid.*, **177**, 237 (1949).

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CATALYTIC DECOMPOSITION OF HYDROGEN PEROXIDE OVER SUPPORTED OXIDES OF MANGANESE

Sir:

The following results are presented in the form of a communication because they apparently offer a clue to a mechanism of heterogeneous catalysis.

Hydrogen peroxide was decomposed over a series of catalysts consisting of oxides of manganese supported on high area γ-alumina. These catalyst samples were prepared by impregnation of alumina with manganous nitrate solution followed by drying and ignition at 200°. The samples differed in percentage of manganese present, and were identical with several samples for which preparation, analytical data, and magnetic measurements have previously been reported.[1]

In each run the weight of catalyst chosen was such that the weight of manganese present was 4.60 mg. The quantity of hydrogen peroxide chosen was 2.00 cc. of 1.14 *M* solution added to the catalyst suspended in 20 cc. of distilled water. The reaction mixture was vigorously stirred, and the rate of oxygen evolution was measured on a flow-meter.

Catalytic activity results are shown in Fig. 1 where the rates of oxygen evolved are plotted against manganese concentration in the several samples.

(1) Selwood, Moore, Ellis and Wethington, THIS JOURNAL, **71**, 698 (1949).