On the Question of a Cysteinyl-glycinase Activity of RNA
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Binkley reported that cysteintyl-glycinase should be a protein-free RNA and put forward the hypothesis that RNA should interfere in protein synthesis through this activity.

Because of the remarkable implications of these statements (among others, that an enzyme should be non-protein in nature), an attempt was made at purifying further the cysteinyl-glycinase (through chromatography on calcium phosphate, Dowex 2 and DEAE cellulose) and at studying some of its chemical properties.

A separation of cysteinyl-glycinase activity from RNA was achieved, and Folin-positive, orcinol-negative, P-negative, chloroform-sensitive preparations of enzyme were obtained, showing a maximum in UV absorption spectrum at about 280 μm.


Glucose Inhibition of Respiration in Ascites Tumor Cells
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Glucose exerts a pronounced inhibitory effect upon the respiration of ascites tumor cells, a phenomenon also called the Crabtree effect. In an attempt to approach this problem the glycolysis and the tricarboxylic acid cycle of the ascites tumor cells were studied under various conditions.

Ascites cells (30–40 mg protein) incubated in phosphate buffer for 1 h at 37°C with excess glucose as substrate were found to accumulate 1–2 μM of a barium insoluble fructose ester, assumed to be fructose-diphosphate.

In the presence of glucose (10 μM/ml), the removal of inorganic phosphate (20 μM/ml) from the medium increased the oxygen uptake by 25–50% and decreased the glucose uptake.


A Method for the Isolation of 2-Deoxy-D-ribose from Thymidine
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It has always been assumed that the sugar component of thymidine is in analogy with the purine nucleosides 2-deoxy-D-ribose. Good evidence in support of this assumption has recently been provided, although the conclusive chemical proof was lacking. The present work describes the isolation of crystalline 2-deoxy-D-ribose from thymidine, and also provides a very convenient micro-method for the isolation of this sugar for tracer studies, which was the main purpose of this work. Thymidine is reduced by 3% NaHg in aqueous solution at room temperature. The reduction mixture was shown by paper electrophoresis to contain two acidic 2-deoxy-D-ribose components formed by breakage of the reduced pyrimidine ring under the prevailing alkaline conditions. Removal of alkali in the reduction mixture with Amberlite IR 120(H) followed by gentle acid hydrolysis with dilute sulphuric acid and passage through a column of weakly basic resin such as Amberlite IR 4B(OH) afforded crystalline 2-deoxy-D-ribose. The yield was almost 100%. For tracer work it is advisable to purify it by recrystallization or paper chromatography.

2. Whittaker, V. F. Physiol. Rev. 31 (1951) 312.
as well as the lactic acid formation by 30–50%, while the blank respiration was unaffected. In this experiment the cells were incubated in trisbuffer with and without phosphate, and special care was taken to maintain the same ionic concentrations of sodium and potassium. The oxygen uptake was measured in a Warburg apparatus for 1 h at 37°. This experiment is in apparent contrast to the results reported by Ibsen, Mc Carty and Mc Kee1. These authors operated with glucose concentrations of less than 3 μM/ml and measured the oxygen uptake during a 3 h period. The small glucose concentrations employed may explain the discrepancy, because in the presence of phosphate (20 μM/ml) more than twice as much glucose was taken up after 1 h by the cells (13–15 mg protein) in our experiments.

In iodoacetate (10−4 M) treated cells phosphate on the contrary slightly stimulated the respiration and exerted no appreciable effect upon glucose uptake or lactic acid formation. Increasing amounts of iodoacetate (0—1.5 x 10−4 M final concn.) successively enhanced the oxygen uptake of ascites cells incubated with glucose as substrate in a way fairly parallel to the normalization of the pH, while the blank respiration decreased slightly. Judged from the glucose uptake and the formation of fructose-diphosphate and lactic acid, an excess of adenosinediphosphate is likely to be formed extramitochondrially in IAA poisoned cells. The still existent Crabtree effect may be explained in accordance with Rackers shuttling theory 2 if one presumes a rate difference as to the permeation of adenine nucleotides in and out of the mitochondria.

Dinitrophenol (DNP, 10−4 M) increased the glucose uptake and lactic acid formation markedly. DNP initially stimulated the oxygen uptake more than the blank when glucose was substrate, but fell off earlier. Excess fumarate completely abolished the glucose inhibition of the respiration in malonate treated cells. The oxygen uptake was also unaffected by malonate in the presence of glucose and fumarate. Glucose did not appreciably affect the citrate formation from fumarate in these cells. Glucose increased the concentration of α-ketoglutarate and reduced the formation of succinate when the cells were incubated in the presence of excess citrate and malonate. Under similar conditions glucose markedly inhibited the oxygen uptake in the presence of excess citrate or α-ketoglutarate.

Hence there was found no evidence for an inhibition of the tricarboxylic acid cycle between fumarate and citrate by glucose. The results may be explained by assuming a partial block of the cycle between α-ketoglutarate and succinate and that this step is rate limiting of the overall mitochondrial respiration in the presence of glucose.


Estimation of Corticosteroid Metabolites in Urine
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During the last decade the importance of adequate methods for evaluation of the adrenocortical function has been more clearly recognized. We have compared the methods most used for corticosteroid determinations in urine and related them to the structure of the steroid hormones and their metabolites. We have worked especially with the method developed by Norymberski and coworkers which we think is superior to other methods 3,4. This method converts, by an oxidation with sodium bismuthate, corticosteroids of certain types called 17-ketogenic steroids (17-KGS) to 17-keto steroids (17-KS). Apart from the desired oxidative effect of the sodium bismuthate on the side chain at carbon atom 17, other effects appear which can cause faulty results. That is especially the case with dehydroepiandrosterone, which is almost completely transformed to other products by the combined effects of sodium bismuthate and acid hydrolysis. The bismuthate probably causes an oxidation to the 5,6-epoxide which is then further rearranged by the acid hydrolysis. This deleterious effect can be minimised by using short oxidation time and it can be completely avoided by using the modification proposed by Appleby et al.5

A system has been worked out for the separation and estimation of the different more important corticosteroid metabolites in urine. It employs enzymatic hydrolysis of the corticosteroid conjugates with β-glucuronidase, oxidation of corticosteroids to 17-KS with sodium bismuthate, reduction of ketonic groups to

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