With the D-amino acid oxidase test it was shown that flavin-adenine dinucleotide is the prosthetic flavin of Y_1, Y_2 and ETF**. Small amounts of iron were consistently found in Y_1 and ETF at a ratio of one iron to six or more flavins. The significance of this small amount of iron is not clear. No other metals were found in the flavoproteins at a significant level, except for copper in G. In view of the findings reported above the actual role of copper in G will have to be reinvestigated.

G, Y_1 and Y_2 can be separated and prepared in high purity from an extract of mitochondrial acetone powder in one procedure involving only seven steps. ETF of high purity is obtained in a separate four step procedure from the same source. Details of these procedures, spectral characteristics and criteria of purity of the new flavoproteins will be reported elsewhere.

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FREDERICK L. CRANE***
JENS G. HAUGE***
HELMUT BEINERT

Institute for Enzyme Research, University of Wisconsin, Madison, Wisconsin (U.S.A.)

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** Similar results had been reported earlier for G from beef liver[6,7].
*** Post-doctoral trainee of the National Heart Institute of the National Institutes of Health.

The occurrence of a direct oxidative pathway of carbohydrate metabolism in the fly *Musca domestica* L.

The carbohydrate metabolism of Diptera has been shown to be similar to that found in other animals. In this group of insects, the oxidation of glucose to CO_2 and water is mediated by the universally accepted glycolytic scheme[1] and citric acid cycle[2]. An investigation of the metabolism of the flight muscles of the housefly has revealed the presence of another route by which glucose may be metabolized. This alternative direct oxidative pathway appears to be essentially similar to that operating in higher animals[3-6] and plants[7].

The enzymic activities of glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase were assayed spectrophotometrically according to GLOCK AND MCLEAN[6]. The reactions proceed as follows:

$$\text{(1) Glucose-6-phosphate} + \text{TPN} \xrightarrow[\text{dehydrogenase}]{\text{Glucose-6-phosphate}} \text{6-phosphogluconate} + \text{TPNH}_2$$

$$\text{(2) 6-phosphogluconate} + \text{TPN} \xrightarrow[\text{dehydrogenase}]{\text{6-phosphogluconate}} \text{ribulose-5-phosphate} + \text{CO}_2 + \text{TPNH}_2$$

Fig. 1 indicates that both dehydrogenases were active in the presence of their respective substrates. When to the system was added lactic dehydrogenase which is capable of reacting with $TPNH_2$[8] pyruvate was rapidly reduced due to the oxidation of $TPNH_2$. It can be seen that glucose-6-phosphate dehydrogenase is approximately twice as active as 6-phosphogluconate dehydrogenase. Both enzymes were specific for TPN.

The reaction mixture with 6-phosphogluconate as substrate accumulated a product which reacted positively on the orcinol test and was probably a pentose.

The breakdown of ribose-5-phosphate to sedoheptulose-7-phosphate is catalyzed by pentose phosphate isomerase and transketolase[3,4]. Sedoheptulose-7-phosphate is then converted to hexose monophosphate by transaldolase[5]. The activity of transketolase was measured according to HORECKER *et al.*[4]. The reaction proceeds as follows:

(3) Ribulose-5-phosphate $\longrightarrow$ Triosephosphate + "active glycolaldehyde"

(4) Triosephosphate + $DPNH_2 \longrightarrow$ α-glycerophosphate + DPN

Overall reaction:

Ribulose-5-phosphate + $DPNH_2 \longrightarrow$ α-glycerophosphate + DPN + "active glycolaldehyde"

Because these insects possess an active α-glycerophosphate dehydrogenase[1], $DPNH_2$ should be oxidized if triosephosphate is a product of pentose breakdown. The data in Fig. 2 indicate an active transketolase as measured by the rapid oxidation of $DPNH_2$ in the presence of ribose-5-phosphate which was probably first converted to ribulose-5-phosphate by pentose phosphate isomerase.

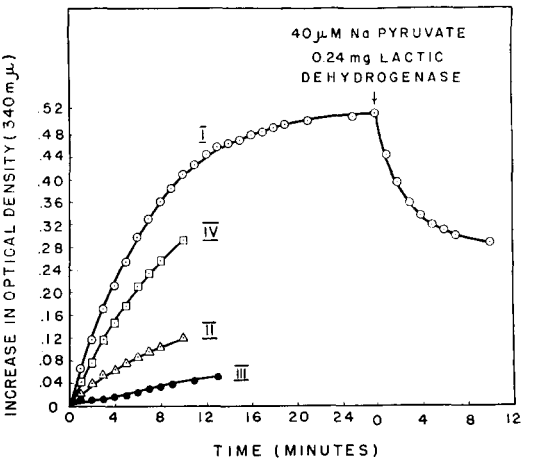

Fig. 1. Glucose-6-phosphate and 6-phosphogluconate dehydrogenase activities of housefly muscle homogenate. The reaction cuvette contained 60 μM of $MgCl_2$, 0.26 μM of TPN, 450 μM of glycylglycine buffer pH 7.6, 0.4 ml of housefly homogenate (2.9 mg dry wt) and in: I. 13 μM of G-6-P and 9 μM 6-P-G; II. 9 μM 6-P-G; III. No substrate; IV. Glucose-6-phosphate dehydrogenase corrected for 6-phosphogluconate dehydrogenase activity (I–II). Total volume was 3.0 ml. The reaction was run at 25° C and measurements were made in a Beckman DU spectrophotometer. The tissue was prepared by homogenizing 50 thoraces of 3-day old female flies for 1 minutes in cold 0.9% KCl solution, filtering through muslin and centrifuging at 3500 g for 10 minutes. The clear supernatant was used.

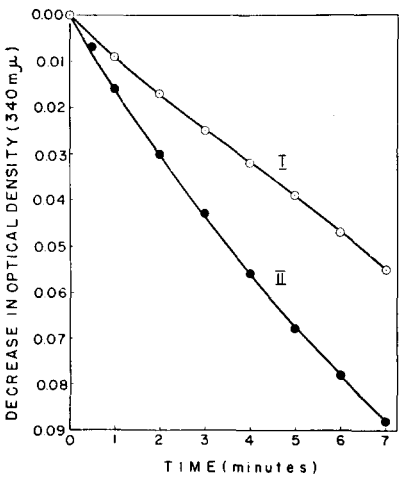

TABLE I

STOICHIOMETRY OF RIBOSE-5-PHOSPHATE CONVERSION

The reaction mixture contained 9 μM ribose-5-phosphate, 76 μM of glycylglycine buffer pH 7.4, 300 μM of $MgCl_2$, 3.0 ml of tissue (21.6 mg dry wt) and enough water to make total volume of 15.0 ml. The incubation period was 80 min at 25° C. All values were corrected for endogenous activity. The homogenate was prepared in distilled water as already described omitting the centrifugation. Fifty thoraces from newly emerged flies were used.

Ribose-5-phosphate consumed	6.0 μM
Triose formed	1.8 μM
Heptulose formed	1.3 μM
Hexose formed	2.8 μM

Fig. 2. Transketolase activity of housefly muscle homogenate. The reaction cuvette (curve II) contained 20 μM of $MgCl_2$, 6 μM of cysteine hydrochloride, 0.4 μM of thiamine pyrophosphate, 0.3 μM $DPNH_2$, 12.5 μM of ribose-5-phosphate, 192 μM of glycylglycine buffer pH 7.6, 1 ml of homogenate (1.6 mg thoraces/ml). Blank cuvette (curve I) contained no substrate. The tissue was prepared by homogenizing 10 thoraces from 3-day old female flies in 50 ml of ice cold 0.9% KCl solution for 1 minute, filtering through muslin and centrifuging at 3500 g for 10 minutes. The reaction was run at 25° C and measurements were made in a Beckman DU spectrophotometer.

The occurrence of triose in this reaction mixture, as verified by chemical analysis, is shown in Table I. Heptulose and hexose were also present. The large amounts of hexose suggests that heptulose was rapidly converted to hexose by transaldolase. The stoichiometry of the reaction indicates that the sum of triose, hexose and heptulose appearing was 5.9 μM as compared with 6.0 μM of pentose disappearing. Hence it appears that in housefly muscle homogenate two pentose

phosphate molecules are converted to heptulose and triose phosphates. The heptulose phosphate can react further to form hexose phosphates.

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W. CHEFURKA

Contribution No. 52, Science Service Laboratory, Canada Department of Agriculture, University Sub Post Office, London, Ontario (Canada)

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REVUE DE LIVRE

Glutathione. Proceedings of the Symposium held at Ridgefield, Connecticut, November 1953, edited by S. P. COLOWICK, A. LAZAROW, E. RACKER, D. R. SCHWARZ, E. STADTMAN AND H. WAELSCH, Academic Press, Inc., New York, 1954, 341 pp., 59 figs; bound, $ 7.50.

Le présent volume comporte quatre parties: La première est consacrée à l'étude des propriétés physiques et chimiques du glutathion. Un excellent article de CALVIN discute la constitution chimique des mercaptans et des disulfures; le spectre u.v. du glutathion indique que ce dernier existe, au moins en partie, sous forme thiazoline. Une étude de BENESCH *et al.* montre que dans les dérivés thiol, même relativement simples, le groupe -SH est partiellement impliqué dans un système où son hydrogène forme un pont avec l'azote aminé; ce pont étant détruit par l'urée à forte concentration. WIELAND donne un exposé clair et complet des travaux récents concernant la chimie du glutathion; la réaction avec les composés carbonyl aboutissant à la formation de semimercaptals semble devoir jouer un rôle important.

La deuxième partie concerne les méthodes de dosage et de caractérisation des composés sulfhydrylés. Après une revue des méthodes classiques, due à PATTERSON ET LAZAROW, et une étude de la chromatographie sur papier du glutathion et de ses produits d'hydrolyse, par GUTCHO ET LAUFER, on y rencontre un intéressant exposé de BARRNETT ET SELIGMAN sur les méthodes récemment mises au point pour la détection histochimique des dérivés sulfhydrylés et des disulfures, notamment à l'aide du 2,2'-dihydroxy-6,6'-dinaphthyl disulfure.

La troisième partie concerne plus particulièrement les mécanismes biochimiques auxquels prend part le glutathion. Une excellente vue d'ensemble sur l'oxydation et la réduction enzymatique du glutathion est due à VENNESLAND ET CONN. La biosynthèse du tripeptide est traitée par SNOKE ET BLOCH, et par STRECKER ET STEKOL, et son rôle dans les réactions de transpeptidation est étudié par HANES *et al.*; les réactions de transfert du radical γ-glutamyl font l'objet d'un article de WAELSCH. RACKER discute l'importance du glutathion en tant que coenzyme dans divers systèmes enzymatiques plus ou moins complexes. Le rôle des composés sulfhydrylés comme accepteurs de groupes acyl fait l'objet d'une étude particulièrement intéressante de STADTMAN; l'action des thioltransacetylases y est soulignée; il est assez remarquable qu'une substance aussi simple que H_2S puisse être un excellent accepteur de groupes acyl, alors que la cystéine et le glutathion sont pratiquement inertes à ce point de vue. La question si difficile des relations entre la présence de groupes thiol dans la cellule et la croissance est clairement exposée par MAZIA.

La quatrième partie est consacrée aux aspects cliniques et aux actions physiologiques du glutathion. Quoique le nombre des observations rapportées soit considérable, les données apparaissent ici moins précises, et le rôle du glutathion y est moins nettement dégagé que dans les trois premières parties.

Dans leur ensemble les exposés constituant le présent volume sont d'une haute tenue; de plus chacun est suivi d'une discussion qui se révèle souvent au moins aussi importante que l'exposé lui-même. Une excellente présentation et une abondante bibliographie augmentent encore la valeur d'un ouvrage dont la lecture s'impose à tout scientifique désireux de se maintenir au courant du développement d'un chapitre de la biochimie qui devient de jour en jour plus important.

CL. FROMAGEOT (Paris)