this case the hydrogenated acids consisted of 47 per cent stearic and 53 per cent palmitic. Assuming that the action of the venom phospholipase is at the α′-position, as in the case of the lecithinase *A*, it may be concluded that the phosphatidylethanolamine of egg yolk has the same type of fatty-acid distribution as the lecithin.

A detailed account of this work, which forms part of the programme of the Food Investigation Organization of the Department of Scientific and Industrial Research, will be published elsewhere.

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Characterization of an Enzyme reducing Pyrroline-5-Carboxylate to Proline

Several lines of evidence serve to establish glutamic-γ-semialdehyde as the key intermediate in the pathway of the biosynthesis of proline from either glutamic acid or ornithine[1,2]. Since glutamic-γ-semialdehyde spontaneously cyclizes nearly completely to Δ^1-pyrroline-5-carboxylic acid[2], hydrogenation of the latter becomes a key reaction in the biogenesis of proline.

An enzyme catalysing this reaction has been observed in extracts of mycelial pods of *N. crassa* and named pyrroline-5-carboxylate reductase[3]. Extracts of rat liver mitochondria have been shown to convert glutamic semialdehyde to proline; but efforts to increase the activity failed[4].

We have determined that pyrroline-5-carboxylate reductase occurs in the soluble proteins of liver. The enzyme has now been partially purified by ammonium sulphate fractionation and the hydrogen donor has been shown to be reduced diphosphopyridine nucleotide. The enzyme has also been characterized with respect to a number of its properties.

Assays of the enzyme are carried out by measuring the fall of absorption of reduced diphosphopyridine nucleotide at 340 mμ in the Beckman spectrophotometer as the reduction proceeds (see Table 1).

Table 1

Experiment	Change in optical density per 10 min.
1. Pyrroline-5-carboxylate + enzyme (0·8 mgm.) + DPNH	0·300
2. DPNH + enzyme (no pyrroline-5-carboxylate)	0·01
3. DPNH + pyrroline-5-carboxylate	0

The enzyme is prepared from rat liver homogenized in a Potter–Elvehjem homogenizer with 1·5 volumes of 0·025 *M* phosphate buffer at *p*H 8 and centrifuged at 100,000 *g* for 30 min. Neutral saturated ammonium sulphate is stirred into the supernatant fluid at 4° to obtain the fraction precipitating between 40 and 60 per cent saturation. This is reprecipitated, dissolved, then reprecipitated and subjected to successive elutions with 50, 45, 40 and 35 per cent saturated ammonium sulphate respectively. The solution resulting from elution with 35 per cent saturated ammonium sulphate is usually most active. This is dialysed for 18 hr. at 4° against four changes of 0·05 *M* phosphate buffer, *p*H 8.

The cuvette for assay contains 3 ml. of 0·05 *M* triethanolamine, 0·04 ml. of pyrroline-5-carboxylate, 0·1 ml. of enzyme solution, containing approximately 1 mgm., and 0·2 ml. reduced diphosphopyridine nucleotide (~0·5 mgm.). The total mixture is adjusted to *p*H 6·8.

The crude supernatant fluid contains an inhibitor fraction which is eliminated as the enzyme is purified. A doubling of the total enzyme activity is obtained in the first precipitation of the 40–60 per cent saturated ammonium sulphate fraction. This inhibitor seems to be precipitated at less than 40 per cent saturated ammonium sulphate.

A preliminary determination of the properties of pyrroline-5-carboxylate reductase has shown that it is unstable in acid and has an optimum *p*H at 6·8 in 0·05 *M* triethanol amine.

The enzyme is inhibited by *p*-chloromercuribenzoic acid, mercuric and silver ions at a concentration of 10^{-5} *M*. *BAL* (1-hydroxy-2,3-dithiolpropane) protects the enzyme from this inhibition, indicating the participation of an active SH group in the reaction. Ethylenediaminetetraacetic acid does not affect the enzyme activity, but adenosine triphosphate, at a level of 10^{-4} *M*, inhibits the reaction.

Studies are now in progress to characterize still further the properties of the enzyme and the inhibitor fraction.

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Effect of Reserpine on Serotonin in Rabbit Serum

It is generally accepted that the effect of clinical doses of reserpine on the circulation is central in origin and that such doses have no peripheral effect of any significance. It has recently been shown that reserpine and serotonin have similar effects on the central nervous system, and that relatively large doses of reserpine in dogs markedly increase the urinary secretion of a metabolite of serotonin[1]. It has therefore been suggested that certain actions of reserpine are mediated through the liberation of serotonin from the intestine[2].

We have shown that, in rabbits, relatively small doses of reserpine reduce the concentration of serotonin in the serum. In our experiments rabbit serum was tested on the rat œstrous uterus, and compared with known concentrations of 5-hydroxytryptamine creatinine sulphate. The accuracy of this method is about ± 10 per cent. The effect of the serum was presumed to be due to the serotonin content, as histamine does not affect the rat uterus and acetylcholine is not found in serum. As further proof of this presumption, it was shown that similar contractions, produced by the serum, and by

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