

Apparent Phosphate Ion Effect on Activity of Phosphoprotein Phosphatase

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Phosphoprotein phosphatase activity was lost upon dialysis of the extract (rat liver and erythrocytes). The studies (to be reported in detail elsewhere) show an activating effect of a small amount of phosphate ion (to a final concentration of 0.1 to 0.3 mM PO_4 depending on substrate concentration) whereas higher concentrations are inhibitory, as found earlier¹.
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Effects of Carcinogenic Amines on the Protein and Ribonucleic Acid Metabolism of the Liver

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The incorporation in vitro of [^{14}C]-DL-valine into the proteins of rat liver slices was studied after different periods of preincubation with 2-aminofluorene (AF), 2-acetylaminofluorene (AAF), 7-fluoro-2-aminofluorene (FAF), 7-fluoro-2-acetylmino-fluorene (FAAF), 2-aminonaphthalene (AN) and aniline at varied concentrations. While a marked reduction of the incorporation was caused by the polynuclear amines, no significant inhibition was observed in the case of aniline even at relatively high concentrations (10^{-3} M). Similar experiments were performed with AF and AN, with [^{14}C] adenine as isotopic component. Adenine and guanine were isolated from RNA, and their specific activities were determined. Also in this case a marked inhibition was observed, less strong however than in the [^{14}C] valine experiments. Within the group of polynuclear amines the capacity of inhibition was not in detail correlated to the carcinogenic potency. As a rule AN was at least as inhibitory as AF, although AN is known to exert its carcinogenic effect on other target organs than the liver. FAF had a slightly lower inhibitory effect than AF. An inhibition by AF and FAF was obser-

ved not only in rat liver slices but also in liver slices of guinea pig, although this animal is refractory to the carcinogenic action of AF derivatives. Rats were treated *in vivo* with AN (100 mg/kg in 1.0 ml of propylene glycol, *per os*). The incorporation of [^{14}C] L-leucine into proteins by a mitochondria-free liver preparation was studied 4 h after the AN-treatment. The activity of the preparation for leucine incorporation *in vitro* was reduced when compared with similar preparations from untreated rats. The microsomes of the system showed a decreased activity in comparison with normal microsomes.

Inhibitor Effects on Light-Induced Phosphorylation. A Comparison between Plant and Bacterial Systems

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Electron transport in mitochondrial oxidative phosphorylation is strongly inhibited by amytal (I), atebirin (II), antimycin A (III) and 2-*n*-heptyl-4-hydroxyquinoline-N-oxide (IV). The effects of these inhibitors were tested on light-induced phosphorylation (LIP) in washed spinach chloroplasts, and compared with earlier results obtained with washed bacterial (*Rhodospirillum rubrum*) chromatophores¹⁻⁴. The following results were obtained in an ascorbate containing medium, whether or not flavin mononucleotide had been added (aerobic conditions, saturating light intensity, 20°C). A 2×10^{-5} M concentration of I gave only little inhibition, as in bacterial LIP⁴. 5×10^{-5} M concentrations of II, III or IV gave inhibitions between 50 and 100%. The inhibition with II occurs in the same inhibitor concentration region as with chromatophores and indicates participation of chloroplast flavin in the electron transport in chloroplast LIP. For III and IV, however, the concentrations needed are about 1 000-fold higher than with chromatophores (or animal mitochondria).

Earlier results with these inhibitors have indicated that electron transport in bacterial (*R. rubrum*) LIP proceeds over a chain with at least two electron carriers^{3,4}. The present results will be discussed in relation to current schemes of electron transport in plant LIP, and the electron transport chains in plant and bacterial LIP will be compared.

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2. Geller, D.M. *VIIth International Congress for Microbiology, Abstracts*, Stockholm 1958, p. 73.
3. Baltscheffsky, H. *Biochim. et Biophys. Acta.* **40** (1960) 1.
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Diphosphothiamine Disulfide in Baker's Yeast

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About 15 years ago Myrbäck and coworkers¹ indicated that baker's yeast, produced in vigorously aerated conditions, contains nearly all its thiamine as diphosphothiamine disulfide. His findings were met with criticism and his viewpoint was not generally accepted, also owing to the fact that the physiological function of cocarboxylase disulfide has not been conclusively verified.

We have now, in contrary to our earlier attempts² been able to confirm the appearance of diphosphothiamine disulfide in aerobically cultivated baker's yeast. We first repeated the experiments of Myrbäck and found a thiamine derivative in baker's yeast, which after cystein reduction acted like cocarboxylase in the thiochrome reaction. With ionexchange chromatography, using Dowex 1-X resin in formiate form, a thiamine derivative was separated from the perchloric acid extract of the commercial stage of baker's yeast, which was eluted by gradient elution with formic acid or with ammonium formiate considerably later than thiamine, thiamine disulfide or thiamine monophosphate and cocarboxylase, the speed corresponding to that of synthetic diphosphothiamine disulfide (Merck). When oxidized after cystein reduction it gives a thiochrome reaction corresponding to that of cocarboxylase.

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Changes in the Microbial Amino Acid Metabolism Induced by 4-Deoxypyridoxine

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Normal strains of *E.coli* were grown in media containing 4-deoxypyridoxine in amounts of 10, 20, 30 and 40 mg per 10 ml of the culture media. Asparagine was the only source of nitrogen, and the incubation time was 7 to 8 days at 37 C°. At the end of the incubation time the media were centrifuged. The clear supernatants were treated with Amberlite IR 120 resins in acid form and eluted with 1 N ammonium hydroxide. The analyses of the amino acids were performed with paper chromatography. The most characteristic finding was the disappearance of the spots of both glutamic and γ -amino-butyric acids when 4-deoxypyridoxine was present in the culture media. The effect of 4-deoxypyridoxine on the growth was also noticed.

Two Unusual U.V. Absorbing Substances Associated with Amino Acids in Bacterial Extracts

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During an investigation of the nucleotides in various extracts from different bacteria we have detected unusual ultraviolet absorbing acidic substances associated with amino acids.

Crude acid-soluble extracts were made by cold perchloric acid treatment of bacteria disrupted in a bacterial press.

Crude water-soluble extracts were obtained by heating living cells in boiling water for 10 min.

The bacteria were harvested during rapid growth. In some cases the extracted material was adsorbed on Norite and again eluted with 50 % ethanol containing 2 % of conc. ammonia. The extracts were fractionated on a Dowex