nine production from homocysteine and serine by extracts of *E. coli* 3/62, a methionine auxotroph specifically lacking the reductase (Guest, Foster & Woods, 1964). Either H_4PteGlu or $\text{H}_4\text{PteGlu}_3$ serves as cofactor for this process.

Similarly *Coprinus* extracts also stimulate methionine synthesis by extracts of *E. coli* 121/176, another methionine auxotroph, specifically lacking N^5 -methyltetrahydropteroyltriglutamate-homocysteine methyltransferase (Guest, Friedman, Foster, Tejerina & Woods, 1964). Here $\text{H}_4\text{PteGlu}_3$, but not H_4PteGlu , is an effective coenzyme. Like *E. coli*, therefore, *Coprinus* has a methyltransferase specific for conjugated N^5 -methyltetrahydrofolates. This was confirmed by direct assay with $N^5[^{14}\text{C}]$ -methyl- $\text{H}_4\text{PteGlu}_3$ and $N^5[^{14}\text{C}]$ -methyl- H_4PteGlu . Only the former converts homocysteine into methionine by transmethylation.

However, extracts of *E. coli* grown with added vitamin B_{12} can use N^5 -methyl- H_4PteGlu as a methyl donor in a cobalamin-dependent transmethylation to homocysteine; the same enzyme will also use methylcobalamin as a methyl donor (Woods *et al.* 1964). Even when *Coprinus* is grown with added vitamin B_{12} neither of these activities can be detected in extracts. Further, *Coprinus* contains only traces of endogenous vitamin B_{12} .

Coprinus extracts do not use *S*-adenosylmethionine as a methyl donor to homocysteine, nor does *S*-adenosylmethionine dilute the transfer of radioactivity from $N^5[^{14}\text{C}]$ -methyl- $\text{H}_4\text{PteGlu}_3$ to homocysteine.

We conclude that in *Coprinus* the conversion of homocysteine and serine into methionine is similar to the cobalamin-independent pathway in *E. coli*.

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A Reactive Histidine Residue at the Active Site of Pig Heart Mitochondrial Malate Dehydrogenase

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Pig heart mitochondrial malate dehydrogenase (EC 1.1.1.37) was inactivated by iodoacetamide (3mM). By using radioactive iodoacetamide it was found that 2mol of the reagent were incorporated/mol of enzyme (mol.wt. 70000). The reaction occurred with N-3 of a histidine residue, and from the pH-rate profile pK_a 7.1 was determined for this residue. The enzyme lost no activity on incubation with 50mM-iodoacetate at pH 7.80.

NADH gave essentially complete protection against inactivation by iodoacetamide; NAD^+ , ADP-ribose and *N*-methylnicotinamide in contrast gave only partial protection. The substrates, oxaloacetate and malate, did not protect the enzyme, but a mixture of NAD^+ and oxaloacetate protected completely, demonstrating the formation of the abortive ternary complex enzyme- NAD^+ -oxaloacetate. Oxaloacetate also enhanced protection by ADP-ribose and complete protection could be obtained with the mixture, again demonstrating formation of a ternary complex. Thus the ADP-ribose moiety of the coenzyme is important in promoting substrate binding, perhaps by a conformational change in the enzyme.

From the protection data and use of different alkylating agents, some tentative conclusions have been drawn about the nature of the active site of mitochondrial malate dehydrogenase.

Comparative Effects of Chemical Modification on Human Alkaline Phosphatases

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Human alkaline phosphatases (EC 3.1.3.1) can be divided into at least three classes (see Moss, 1969), but, with the exception of placental phosphatase for which amino acid analyses have been published (Ghosh & Fishman, 1968; Harkness 1968), little is known about the underlying structures of these enzymes. Acetylation of rat intestinal phosphatase (Fishman & Ghosh, 1967) and bovine brain phosphatase (Brunel, Cathala & Saintot, 1969), and treatment of kidney phosphatase with cyanate (Carey & Butterworth, 1969), increases the electrophoretic mobility of the enzymes while decreasing their activity. These treatments have been applied to human hepatic, intestinal and