

On the Reactivity of Thiol Groups in Ox Heart Lactic Dehydrogenase

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Lactic dehydrogenase (LDH) is not affected by several common sulfhydryl reagents and is just slowly inactivated by *p*-chloromercuribenzoate (PCMB)¹. In contrast, silver and mercuri ions have been found to inactivate the enzyme very rapidly. The inhibition was counteracted by cysteine. Amperometric titration of the native LDH with mercuri chloride showed the presence of 7—8 thiol groups.

Both the rate and the extent of the reaction with PCMB were increased after acetylation of

LDH with acetic anhydride and after denaturation of the protein with lauryl sulfate.

Iodine was found rapidly to oxidize essential thiol groups. The inactivation was reversed by cysteine. No substitution of tyrosine took place at pH 7. In contrast to iodine, the strong oxidizing agent periodate inactivated LDH very slowly.

The facts that mercuri, silver and iodine were found to oxidize essential thiol groups rapidly, whereas PCMB and periodate caused much slower inhibition, could be due to steric hindrances of the larger molecules. Since the substrates and the coenzyme are comparatively large molecules, thiol groups may not be of direct importance in the binding of coenzyme or substrate in LDH.

1. Neilands, J. B. *J. Biol. Chem.* 208 (1954) 225.