

things, of interest from a nutritional point of view, since stomachs, intestines, and udders are used by some food manufacturers either alone or mixed with other ingredients in various chopped meat products.

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The Solubility and the Salt Sensitivity of Yeast D- and L-Lactic Cytochrome c Reductase

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Homogenates of aerobic yeast have been found to contain D- and L-lactic cytochrome c reductase in the ratio 1:2^{1,2}. Using our extraction method / (15–30°), mainly the L-enzyme was solubilized. At low temperatures (0–5°) both enzymes were obtained in the particulate fraction. Half or more of the L-activity was readily extracted from the particles at 30° with 0.2 M disodium phosphate, whereas the D-enzyme remained insoluble. The extracted particles contained D- and L-lactic cytochrome c reductase in the ratio 1:1. Further mechanical desintegration with ballotini beads gave rise to soluble D-lactic cytochrome c reductase; very little of the L-enzyme was obtained. Complete extraction was not achieved for any of the two enzymes. It is concluded that one part of the L-activity was more firmly bound to the particulate fraction than the other.

Following the described method³, three fractions of lactic cytochrome c reductase were obtained^{2,4}. Fractions I, II, and III were eluted in that order from a N,N-diethylaminoethyl-cellulose column. Fractions I and II were specific for L-lactic acid. Fraction III was D-specific (D-LDH III).

However, small amounts of a L-specific enzyme has now been found in some of the fractions III (L-LDH III).

The D- and the L-enzyme of the cell homogenate were both salt sensitive. The D-enzyme was more salt sensitive than the isolated D-LDH III². The L-enzyme was inhibited 50 % when the phosphate concentration (pH 7.1) was increased from 0.01 to 0.08 μ .

The activity of fractions I and II, which represented most of the L-LDH activity of the purified solution, was quite the same in phosphate of ionic strength 0.01 and 0.08. In contrast, both D- and L-LDH III were inhibited 70–90 % when the buffer concentration was increased from μ 0.01 to 0.08. The inhibition of L-LDH III was the same with low and high concentrations of cytochrome c, whereas the inhibition of D-LDH III was less at high concentrations of the acceptor².

The strong effect of salt on L-LDH of the cell homogenate suggests that fair amounts of the salt sensitive enzyme is present in the cell. In this connection it is interesting that L-LDH obtained by the method of Boeri *et al.*⁶ is very salt sensitive.

To the authors' knowledge, the effect of salts on D- and L-LDH III is stronger than for any other enzymes described. From other relationships D-lactic cytochrome c reductase has been proposed as intermediate in the formation of the L-enzyme^{1,5}.

D- and L-LDH III were eluted together with ribonucleic acids⁷. Furthermore, the enzymes precipitated together with nucleic acids when the eluate was dialyzed against water. A charge aggregate between protein and nucleic acid might explain both the salt sensitivity and the high negative charge which these two enzymes have available to the anion exchanger.

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