Cellular Localisation and Specificity of Intestinal Invertase

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In homogenates from intestinal mucosa (swine), invertase has been found exclusively localized in the microsome fraction, obtained by differential centrifugation. By treating the microsomes with a solution of desoxycholate the invertase activity was obtained in a clear solution which, however, did not prove to be a true solution, as dialysis lead to a recovery of the enzyme in the particulate form.

When isolated microsomes were digested by trypsin, invertase was rapidly inactivated parallel with the hydrolysis of proteins. In the beginning of this reaction, a solubilisation of invertase was observed. Phosphate buffer was found to protect invertase against the inactivation by trypsin, without affecting its solubilisation. In the presence of desoxycholate, it is therefore possible to use trypsin digestion for preparation of soluble invertase.

The relation of invertase to other α-glucosidases of intestinal mucosa is not known. According to the theory of Weidenhagen [1], the animal intestine should contain only one α-glucosidase, hydrolyzing maltose, sucrose, and other α-D-glucopyranosides. Enzyme preparations from animal intestine have, however, no action upon α-methylglucoside [2] and recently two α-glucosides have been demonstrated to be hydrolyzed by enzymes different from maltase, namely *iso*maltose and nigerose [3,4].

During the further purification of solubilized invertase, we have found that the trehalase activity is rapidly lost during ethanol fractionation while invertase and maltase activity are not affected during this procedure. Intestinal trehalase must therefore be an enzyme separate form maltase and invertase.

The further separation of invertase and maltase activity is explored by the use of DEAE-cellulose columns and these results will be discussed.

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3. Larner, J. and McNickle, C. M. *J. Biol. Chem.* **215** (1955) 723.
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Fractionation of Enzymes from Ringhals Venom

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Venom from the South African ringhals cobra *(Hemachatus haemachates)* has been studied by anion exchange chromatography and column electrophoresis.

Early results showed considerable differences in the chromatograms for different batches of venom and it was found that storage and autoincubation of the venom would alter the chromatographic behaviour of especially the phosphodiesterase. These results indicated that the procedure for the collection of the venom might be of significant importance for a successful fractionation. We have later by the kind cooperation of Dr. J. H. Mason at The South African Institute for Medical Research, been able to compare venom collected in the ordinary way (dried at room temperature) and venom "milked" down in a container cooled by dry ice and immediately freeze-dried. The chromatograms of these two materials showed that the main part of the phosphodiesterase is present in different forms depending on the collection procedure and that chromatography of the freeze-dried venom gives about 7 times better purification for the phosphodiesterase than obtained for venom collected in the ordinary way. It has also been found that the successful chromatographic separation of the diesterase is dependent on a sort of double fronting of the developing agent. The phosphodiesterase was separated from lecithinase A and cholinesterase, but the chromatography so far used did not separate the two latter enzymes from each other. However, column electrophoresis at pH 5.5 effects a complete separation of cholinesterase from both lecithinase A and phosphodiesterase. It thus appears that a combination of chromatography and electrophoresis may offer possibilities for an efficient purification of all three enzymes.

Recent Studies on Sialic Acid

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