The optimal hydrogen-ion activity for pancreatic amylase is dependent both upon the kind and concentration of salt present.

In the presence of each of the salts investigated here, the optimal hydrogen-ion activity for pancreatic amylase decreases with increasing concentration of the salt up to a certain salt concentration, beyond which it ceases to be appreciably influenced.

The concentration of salt at which the optimal hydrogen-ion activity for the enzymic activity ceases to be appreciably influenced depends on the salt.

In the presence of more than one of these salts the optimal hydrogenion activity depends upon the concentration of the salts present.

Sodium sulfate and sodium phosphate were found to be without influence on the activity of pancreatic amylase.

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[Contribution from the Department of Chemistry, Columbia University, No. 579]

THE INFLUENCE OF CONCENTRATION OF NEUTRAL SALT ON THE ACTIVATION OF PANCREATIC AMYLASE

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In a previous paper² the optimal hydrogen-ion activity for pancreatic amylase in the presence of different concentrations of each of a series of neutral salts has been reported. These having been established, it was possible to continue the investigation and to determine the optimal concentration of each salt for the activity of the enzyme. The results of this phase of the work will be reported briefly in this paper.

Experimental

A series of parallel hydrolyses of starch by pancreatic amylase was carried out in the presence of different concentrations of each salt with the hydrogen-ion activity in each case adjusted to that previously found to be optimal for the enzyme in the presence of the concentration of salt being studied. The conditions for making up solutions and measuring enzymic activity as there described were maintained throughout. Fig. 1 shows the results obtained from a direct comparison of the activity of pancreatic amylase in the presence of from 0.005 to 0.10 M sodium chloride. The enzyme was found to exert its optimal activity in the presence of from 0.02 to 0.05 M sodium chloride.

With potassium chloride the concentrations investigated were 0.01,

- ¹ We are greatly indebted to the Carnegie Institution of Washington for grants in aid of this investigation.
 - ² Sherman, Caldwell and Adams, This Journal, 50, 2529 (1928).

0.02, 0.03 and 0.05 M, and 0.03 to 0.05 M potassium chloride solutions were found to permit optimal enzymic activity. From a direct comparison of the activity of pancreatic amylase in the presence of 0.01, 0.03, 0.05, 0.10 and 0.20 M sodium bromide it was found that 0.03 to 0.20 M was the most favorable concentration.

Measurements of the activity of pancreatic amylase in the presence of 0.03, 0.05, 0.10, 0.15 and 0.20 M sodium nitrate showed that 0.10 to 0.20 M was the most favorable concentration of sodium nitrate.

The results obtained in the presence of 0.05, 0.10, 0.15 and 0.20 M sodium chlorate solutions were similar to those obtained with sodium nitrate.

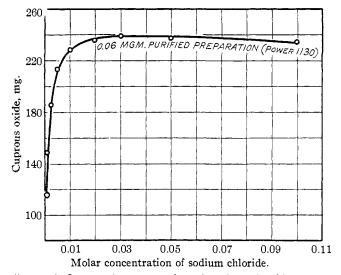


Fig. 1.—Influence of concentration of sodium chloride on the activity of pancreatic amylase.

Comparisons of the activity of pancreatic amylase in the presence of 0.10, 0.15, 0.175 and 0.20 M sodium sulfocyanate or in the presence of 0.10, 0.15, 0.175, 0.20, 0.25 and 0.30 M sodium fluoride showed that a concentration of from 0.15 to 0.20 M sodium sulfocyanate or 0.20 to 0.30 M sodium fluoride was sufficient for the maximum activation which these salts are capable of producing.

An examination of the data summarized in Table I shows that the concentration of salt necessary for maximum activation of pancreatic amylase depends upon the individual salt present.

There is an increase in the enzymic activity as the concentration of salt is increased up to a maximum activity of the enzyme in the presence of each salt. Within the limits of concentration investigated here, further increases in the concentration of salt had practically no influence on the

Table I

The Optimal Concentrations of Certain Salts with the Corresponding Optimal Hydrogen-Ion Activities for the Activity of Pancreatic Amylase^a

Salt	Optimal concn., M	Optimal PH
Sodium chloride	$0.02 - 0.05^{b}$	7.1 - 7.2
Potassium chloride	.0305	7.1 - 7.2
Sodium bromide	.0320	7.1
Sodium nitrate	.1020	7.1
Sodium chlorate	.1020	6.9 - 7.1
Sodium sulfocyanate	. 15- . 2 0	6.7 - 6.8
Sodium fluoride	.2030	6.7 - 6.8

 $[^]a$ Mixtures of acid and alkaline sodium phosphates corresponding to a total concentration of $0.01\ M$ phosphate were present in all cases.

activity of the enzyme or on the hydrogen-ion activity which is optimal for the enzyme in its presence. In the presence of insufficient salt to produce maximum activation, as the concentration of salt is decreased, the enzyme exerts its optimal activity in increasingly more acid solutions. Pancreatic amylase exerts its optimal activity in more acid solutions in the presence of those salts which require higher concentrations for complete activation. All of these results emphasize again the need of carefully standardizing and controlling the conditions for measuring enzymic activity.

Summary

The concentration of sodium or potassium chloride or of sodium bromide, nitrate, chlorate, sulfocyanate or fluoride most favorable (in each case) to the activation of pancreatic amylase has been quantitatively established.

The optimal concentration (in terms of molarity) is found to be different for different neutral salts.

The activity of pancreatic amylase is favored by more acid solutions in the presence of those salts of which higher concentrations are required for its complete activation.

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^b The practically constant activity obtained within the limits of concentration considered here would indicate that there is enough of the salt present fully to activate the enzyme in each case.