

summarised in Table 1. It is notable that crystalline salts were obtained in all cases except three. Also it may be pointed out that most of the acids used in these experiments have rather small tendencies to yield crystalline salts with the common alkaloids. The acids liberated from the first fraction of the (+)-amine salts were obtained in very different degrees of optical purity. In two cases no or very small activity could be detected while in three cases the acids were obtained in a fairly high degree of optical purity. In the remaining six cases the products contained from 10 to 30 % excess of active acid which in many cases is enough to render a resolution possible by repeated recrystallisations. In view of this the use of 2-amino-1-phenylpropane for optical resolution of acids seems to be of great value as a complement to the common alkaloids and as a substitute for  $\alpha$ -phenylethylamine.

The author is much indebted to *AB Astra* for a supply of benzedrine, which was used for the preparation of the optically active amine.

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Received March 31, 1953.

## Volume Changes Accompanying Enzymatic Reactions with Ribonuclease and Deoxyribonuclease

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Previous investigations by Chantrenne, Linderström-Lang and Vandendriessche<sup>1</sup> and Vandendriessche<sup>2</sup> have shown that the enzymatic breakdown of ribose nucleic acid (RNA) by ribonuclease is accompanied by volume changes that cannot be explained solely in terms of a hydrolytic splitting of an ester bond be-

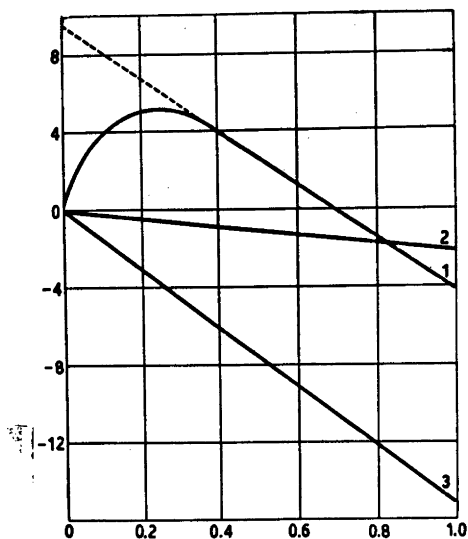


Fig. 1. Relation between volume changes and titration values (pH 4.6—30° C). Absc.: Base consumption in equivalents per mole ( $P_4$  in case of nucleic acids). Ord.: Volume changes in ml per mole (*id.*).

1. Yeast ribose nucleic acid and ribonuclease.
2. *Thymus* deoxyribose nucleic acid and deoxyribonuclease.
3. Ammonium uridine-2'-3'-phosphate and ribonuclease.

tween a phosphate group and a hydroxyl group in ribose. In fact, the splitting of such a bond would produce a decrease in volume of about 2 ml/eq. at pH 4.6, while actually a contraction of 14 ml/eq. is found. Moreover, at certain temperatures (*e.g.* 30° C) this contraction is preceded by a very rapid increase in volume (curve 1, Fig. 1).

Several explanations have been proposed for these phenomena, *viz.* that the dilatation is due to the breakdown of a superstructure, and that the contraction is caused by dipole formation. However, in the light of further experiments at other temperatures and pH-values (Vandendriessche<sup>2</sup>), none of the explanations offered were wholly satisfactory. It has now been found that the enzymatic break-

down of deoxyribonucleic acid (DNA) at pH 4.6 and 30° C is accompanied by a contraction of only 2 ml/eq., and that no initial dilatation occurs (Fig. 1, curve 2). The experiments were made in the way described by Chantrenne, Linderstrøm-Lang and Vandendriessche<sup>1</sup> for the study of RNA-breakdown, *viz.* combined dilatometry and titration of liberated acid groups.

In view of these findings it is natural to correlate the observed difference in behavior between the RNA and the DNA with their main difference in structure, *viz.* the presence or absence of an OH group at position 2'. Since according to Brown and Todd<sup>3</sup> and Brown, Dekker and Todd<sup>4</sup> an intermediate 2'-3' (cyclic) phosphate is formed in the breakdown of RNA, an investigation was made of the volume change accompanying the enzymatic degradation of ammonium uridine-2'-3'-phosphate. As shown in Fig. 1, curve 3, one titratable group is liberated in this substance by the action of ribonuclease and a contraction of 15 ml/eq. is observed. A similar investigation of cytidine-benzyl phosphate b, which according to Brown and Todd should give rise to cytidylic acid and benzyl alcohol with intermediary formation of cytidine-2'-3' (cyclic) phosphate, led to results that may be qualitatively represented by curve 1, Fig. 1. Due to lack of material simultaneous estimations of volume change and release of acid groups could not be carried out. The curve for the relation between volume and time matched however closely those found for RNA showing a rapid initial increase in volume (*ca.* + 4 ml/mole at the maximum) followed by a slower fall. This diphosphate ester therefore behaves almost exactly as RNA, and it is therefore suggested that the initial increase in volume, observed in experiments with RNA and cytidine-benzyl-phosphate b is due to the formation of a 2'-3'-phosphate, while the contraction of about 15 ml/eq.

equally characteristic of the degradation of RNA, uridine-2'-3' phosphate and cytidine-benzyl-phosphate b is due to the opening of an ester bond in the cyclic phosphate. On this basis the experiments on DNA (Fig. 1, curve 2) are easily explained too since the absence of an OH group at position 2' excludes the intermediary formation of cyclic phosphate so that the hydrolysis of the ester bond is uncomplicated and therefore is followed by the normal contraction of about 2 ml/eq.

From these results it would then appear that the formation of cyclic phosphate is accompanied by a dilatation of *ca.* 15-2 = 13 ml/mole, an astonishingly high value for so small a molecular group. It is to be hoped that a theoretical investigation of this phenomenon may throw light upon the structural details of the cyclic phosphates.

The author is indebted to Dr. D. O. Jordan, Dr. J. W. Rowen, and especially to Dr. A. R. Todd for generous gifts of enzyme and substrates, and to Dr. K. Linderstrøm-Lang for constant advice and discussion of the problem.

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Received March 31, 1951.

## Note on the Stepwise Degradation of Peptides *via* Phenyl Thiohydantoins

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A procedure for the stepwise degradation of peptides *via* phenyl thiohydantoins has earlier been described by us<sup>1</sup>. In this