

## Formation of Particulate Ribonucleoprotein with Enzymatic Activities *in vitro*

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During the purification of lactic dehydrogenase from baker's yeast<sup>1</sup> a particulate ribonucleoprotein was observed to be formed in a soluble preparation of proteins and ribonucleic acid. The solution was obtained from the mechanically extracted cells by acetone precipitation, calcium phosphate adsorption/elution and ammonium sulfate precipitation. The nucleoprotein precipitated on storage at 6°C from the saltfree solution which had been concentrated by lyophilization.

The precipitate contained about equal amounts of nucleic acid and protein. Adenine, guanine, cytosine and uracil were all present. The nucleoprotein (nucleic acid and protein) was insoluble in water and in different salt solutions (*e. g.* 1 M sodium chloride), but dissolved in 0.04 M sodium hydroxide. In some preparations the particles settled easily in a low centrifugal field, in others a clear solution was not obtained even after 30 min centrifugation at  $30\,000 \times g$ .

The original solution contained 10 % ribonucleic acid, 5–10 % each of lactic-, malic-, alcohol- and triosephosphate dehydrogenase, and had proteolytic and nucleolytic activities. The well-washed particles had malic dehydrogenase activity. None of the other dehydrogenases were present. The particles digested the particulate ribonucleic acid. Thus, on incubation in phosphate buffer purine and pyrimidine material appeared in the supernatant. This material was not liberated by boiling of the particulate ribonucleoprotein.

Malic dehydrogenase was measured spectrophotometrically at 340 m $\mu$ , with oxalacetic acid and DPNH at pH 8.4, with malic acid and DPN at pH 9.6. The rate was 40  $\mu$ moles oxalacetic acid reduced per mg particulate protein per hour. Highly purified malic dehydrogenase has been obtained from the original ammonium sulfate precipitate by zone electrophoresis on cellulose. This soluble preparation did not contain ribonucleic acid and had an activity of 8 500  $\mu$ moles oxalacetic acid reduced per mg protein per hour.

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The results indicate that particulate ribonucleoproteins isolated from bacterial cells may not necessarily represent original particles of the cell. Particles may be formed in a haphazard manner during extraction and purification.

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## Standard Deviations of Interatomic Distances in Completely Deuterated Benzene

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In a previous communication the calculation of standard deviations of interatomic distances in the benzene molecule has been reported<sup>1</sup>. The calculated values have been compared with values obtained from electron diffraction data<sup>2</sup>.

Similar calculations from spectroscopic data have been performed for the benzene-d<sub>6</sub> molecule with the result given in Table 1. The experimental fundamental

Table 1. Standard deviations for interatomic distances in benzene-d<sub>6</sub>.

Distance	Calculated standard deviations in Å units			
	$T = 0$	273.16	298.16	323.16
C <sub>1</sub> —D <sub>1</sub>	0.0660	0.0660	0.0660	0.0660
C <sub>2</sub> —D <sub>1</sub>	0.0889	0.0895	0.0898	0.0902
C <sub>3</sub> —D <sub>1</sub>	0.0835	0.0845	0.0848	0.0952
C <sub>4</sub> —D <sub>1</sub>	0.0821	0.0834	0.0839	0.0844
C <sub>1</sub> —C <sub>2</sub>	0.0455	0.0456	0.0457	0.0458
C <sub>1</sub> —C <sub>3</sub>	0.0529	0.0539	0.0543	0.0546
C <sub>1</sub> —C <sub>4</sub>	0.0570	0.0587	0.0593	0.0599
D <sub>1</sub> —D <sub>2</sub>	0.1277	0.1290	0.1296	0.1303
D <sub>1</sub> —D <sub>3</sub>	0.1095	0.1107	0.1112	0.1117
D <sub>1</sub> —D <sub>4</sub>	0.1012	0.1024	0.1028	0.1033

frequencies given by Brodersen and Langseth<sup>3</sup> have been applied. To the knowledge of the author, the molecule has not been investigated by electron diffraction.

1. Cyvin, S. J. *Acta Chem. Scand.* **11** (1957) 1499.