

collagen to be the true value for pure collagen. The differences shown here between neutral salt-soluble and citrate-soluble collagen affect the same amino acids as the differences described by BOWES *et al.*⁴ between citrate-soluble and insoluble collagen from bovine skin. These differences were attributed by these authors to the presence of a mucoprotein containing hexosamine in insoluble collagen. However, hexosamine is absent from neutral salt-soluble collagen so that this suggestion would not apply here.

The presence of ornithine in collagen has not previously been reported but its origin is not known.

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Fractionation of ribonucleic acid by precipitation with neutral salts*

Methods of fractionation of ribonucleic acid (RNA) preparations have been based on fractional precipitation or extraction by suitable solvents^{1,2}, elution from ion-exchangers^{3,4}, or dissociation of protamine nucleate⁵. The last method is used for obtaining fractions with different base composition, while the others give fractions with different molecular weight.

High-molecular-weight RNA has been shown to be precipitated from neutral aqueous solution by high concentrations of salts such as NaCl or (NH₄)₂SO₄^{6,7}, or by relatively low concentrations of MgCl₂ or CaCl₂⁸. We have now been able to divide yeast RNA into fractions with different base composition by the fractional precipitation with neutral salts. This method has the advantages of being simple and being applicable to high-molecular-weight RNA preparations and to any size of sample.

RNA was prepared from baker's yeast by the method of CRESTFIELD, *et al.*⁹. It was dissolved in dilute saline, and then an appropriate volume of the concentrated salt solution (NaCl or MgCl₂) was added. The mixture was kept at 0–5° for 15–20 h. The precipitate formed was separated by centrifugation. In this way the sample may be fractionated successively into several fractions. RNA's in the precipitate and in the supernatant were analyzed for their base composition⁹ after being recovered by ethanol. The results are shown in Table I.

It can be seen from Table I that the fractions thus obtained, except those of Expt. 4, differ from each other with respect to their guanine and cytosine contents; the fraction more soluble in salt solution tends to be richer in guanine and cytosine than the one less soluble. Especially the fraction soluble in 2 M NaCl obtained in Expt. 2 had markedly high guanine and cytosine contents. On the other hand, the composition of the readily-precipitable fraction did not vary greatly from the original sample. In Expt. 4, the readily-precipitable fraction was sub-divided into smaller fractions, but no significant variation of the composition could be observed, although it may also be possible that MgCl₂ is less effective for fractionation than NaCl, since even the supernatant fraction in this experiment differed little from the starting material.

From these results it seems possible that there are some RNA molecules which are exceptionally rich in guanine and/or cytosine in yeast RNA preparations obtained by the method of CRESTFIELD *et al.*⁹; they may represent a relatively small fraction and can be concentrated in a fraction

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TABLE I
BASE COMPOSITION OF FRACTIONS SEPARATED FROM YEAST RNA
Expts. 1-3 refer to one preparation, Expt. 4 to another.

Expt. No.	Salt	Fraction	%	Base composition (moles base/mole adenine)				No. of determinations
				Adenine	Guanine	Cytosine	Uracil	
Starting material				1.00	1.02	0.82	1.01	12
1.	NaCl (1.0 M)	Precipitate	68	1.00	1.05	0.77	0.98	5
		Supernatant	32	1.00	1.12	0.86	1.00	5
		Weighted mean §			1.00	1.07	0.81	0.98
2.	NaCl (2.0 M)	Precipitate	85	1.00	1.05	0.76	0.99	6
		Supernatant	15	1.00	1.27	1.10	0.95	7
		Weighted mean §			1.00	1.08	0.81	0.98
3.	MgCl ₂ (0.017 M)	Precipitate	46	1.00	0.99	0.77	1.00	7
		(0.020 M)	Precipitate	18	1.00	1.02	0.82	1.04
	Supernatant		36	1.00	1.08	0.88	1.01	8
		Weighted mean §			1.00	1.03	0.82	1.01
Starting material				1.00	1.03	0.77	1.01	5
4.	MgCl ₂ (0.015 M)	Precipitate	7	1.00	1.01	0.75	1.00	5
		(0.017 M) Precipitate	14	1.00	1.04	0.77	1.01	5
		(0.019 M) Precipitate	18	1.00	1.02	0.76	1.02	5
		(0.024 M) Precipitate	29	1.00	1.01	0.77	0.99	5
		Supernatant	32	1.00	1.03	0.81	0.99	5
		Weighted mean *			1.00	1.02	0.78	1.00

§ Calculated for the total fractions. These values should be compared with those for the corresponding starting material.

soluble in concentrated NaCl. They probably have a relatively low molecular weight (*cf.* ref.⁸). The molecular weight and the chromatographic behavior⁸ of these fractions are now under investigation; the results will be reported elsewhere.

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