

A Study on the Structure of a Viral RNA

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SAXS was applied to the RNA from bacteriophage MS2. From measurements in a 0.1 M NaCl solution containing 0.5 mM MgCl₂ the molecular weight of the RNA was obtained as 1.09×10^6 and the radius of gyration as 176 ± 5 Å. The largest diameter of the RNA particle was determined as 620 Å. Two cross-section factors were derived from the scattering curve. The one at small angles corresponded to a cross-section radius of gyration $R_c = 84.3$ Å and to a mass per unit length $M_c = 1890$ Å⁻¹. From the second cross-section factor, which was found at larger angles, another R_c of 9.1 Å and another M_c of 169 Å⁻¹ were derived. The latter value is smaller than the theoretical mass per unit length for a RNA double helix, but it is definitely larger than the value of 91 Å⁻¹ found for MS2 RNA in 4.6% formaldehyde solution. It appears to be consistent with a double-helix content of about 70%. Comparison of the scattering curve for MS2 RNA with theoretical curves for various models led to the conclusion that the native MS2 RNA is a flat particle, perhaps an elliptic cylinder with axes of 618 Å and 312–331 Å and a height of 55–110 Å. Further experiments with the RNA were performed in 4.6% formaldehyde solution at 60°C. At this temperature the RNA obviously behaves like a random coil, because, besides a cross-section radius of gyration of 7 Å and a mass per unit length of 71 Å⁻¹, a persistence length of 59 Å could also be found.

Small-Angle X-ray Scattering Study on the Recognition of a tRNA Substrate by its tRNA Ligase

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As the first step of a small-angle X-ray scattering study on the interaction of ligases with their specific tRNA substrate, the lysine-tRNA ligase from yeast has been characterized. Small-angle X-ray data yield a radius of gyration equal to 37.5 Å, a molecular weight of 114 000 and a volume of 295 000 Å³. A comparison of the experimental data with the theoretical scattering curves indicates that the shape of the molecule can be represented as an oblate ellipsoid. In the next step solutions containing both ligase and specific tRNA have been studied; the X-ray intensities at small angles increased dramatically compared to the intensities obtained by adding those recorded from separate solutions of ligase and tRNA. However, solutions of ligase and unspecific tRNA did not yield any measurable increases in the intensities. This increase in intensity, ΔI , produced by the specific tRNA, is only a function of the complexes formed between the ligase (L) and its tRNA substrate (S), provided that the intensity constitutes the sum of the intensities for all the particles in the solution. Such ΔI values have been recorded for two sets of solutions where, in each set, the solutions contained varying tRNA concentrations (A), but a constant ligase concentration (B). When normalized data, $(\Delta I)/B$ against A at constant B , for a certain low angle were compared to a series of curves simulated for different complexes of the type $L_p S_q$, it was found that the curves produced by the $L_2 S$ complex yielded the best fit. A further support for the formation of such a complex was indicated through the determination of molecular weight and volume.