The helical arrangement of the protein sub-units
in tobacco mosaic virus

X-ray diffraction studies have established that in the tobacco mosaic virus (TMV) particle the virus protein is in the form of structural sub-units set in helical array about the particle axis, and that there are (very nearly) \(3n+1\) such sub-units on three turns of the helix. The pitch of the helix is 23 Å and the axial repeat period, therefore, 69 Å. The value of \(n\) has, however, remained uncertain. It has been estimated as 10 or 12, giving 31 or 37 sub-units in the axial repeat period. We now know that both of these estimates are false, and believe the correct value of \(n\) to be 16, giving 49 sub-units in 3 turns of the helix.

The method used to establish this result was altogether more rigorous than that used to obtain the earlier estimates. It will be described in detail in *Acta Crystallographica*. It is based on a detailed quantitative comparison of the X-ray scattering by normal TMV and by a mercury-substituted TMV, (Hg-TMV), kindly prepared for us by Dr. Fraenkel-Conrat. In this preparation mercury was bound to the cystein residue of the virus protein, in the form -Hg-CH₂, to the extent of one Hg atom to about 20,000 molecular weight of virus. Since there is only one cystein residue in each chemical sub-unit of virus protein it is to be expected that all mercury atoms occupy equivalent sites in Hg-TMV, and hence that they all lie at the same radial distance from the particle axis. Our X-ray diffraction measurements, on the equator and first five layer-lines of the fibre-diagrams of TMV and Hg-TMV, led us to conclude that all (or very nearly all) the mercury atoms lie on a helix of radius 57 ± 1 Å, and, further, that in the axial repeat period of 69 Å there are 3 turns of the helix and 49 equally spaced mercury sites.

Since there is only one mercury site on each chemical sub-unit, this indicates that there are 49 chemical sub-units on 3 turns of the helix. Using this result, it is found that a self-consistent interpretation of a substantial part of the X-ray diagram of TMV is obtained if it be supposed that
there are 49 structural sub-units in 3 turns of the helix. There is thus strong evidence that the sub-units determined by chemical methods on the one hand, by X-ray methods on the other, are identical.

We must now consider the implications of this conclusion with regard to the relationship between the molecular weights of the sub-unit and of the complete virus particle. Taking the particle length to be 3000 Å, there are 49 × 3000/69 = 2130 sub-units in the complete particle. If the molecular weight of a sub-unit is 17,000, this gives a total molecular weight of 36.7 × 10^6 for the protein or, assuming a nucleic acid content of 6%, a molecular weight of 38.4 × 10^6 for the virus particle. If the sub-unit weight is 18,000, the particle weight calculated in this way is 40.7 × 10^6.

Our results therefore suggest that the molecular weight of TMV is about 40 × 10^6. This is in good agreement with the values obtained by Scheffers and Hergold using ultracentrifugation and diffusion measurements, and by Oster, Doty and Zimm and Oster, using light-scattering measurements. On the other hand Williams, Backus and Steer, using a direct method involving weighing and electron microscope measurements, found the particle weight to be 50 × 10^6.

We do not attempt, here, to decide between these conflicting values. We wish to stress, however, that, as far as the chemical and X-ray determination of the particle weight of TMV is concerned, the value, 40 × 10^6, arrived at above refers to the RNA together with the protein sub-units which follow the helical arrangement described. On the basis of the X-ray data alone we cannot exclude the possibility that the virus particle contains small amounts of protein organised in a different way. The combination of chemical and X-ray data, however, probably provides the most accurate method of determining the weight of that part of the virus which follows the regular helical arrangement.

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Rosalind E. Franklin
K. C. Holmes

Birkbeck College Crystallography Laboratory, London (England)

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