

Small-Angle X-ray Scattering Functions for Ellipsoids of Revolution and Right Circular Cylinders*

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The angular intensity dependence of X-rays scattered at small angles from randomly oriented particles in the colloidal size range has been evaluated numerically for ellipsoids of revolution and right circular cylinders of a wide range of axial ratios. The usefulness of the result with respect to experimental determination of colloidal shapes is discussed. It is shown that, for all the elongated shapes studied, subsidiary maxima which appear in the low-intensity region of the scattering curve are a function of the equatorial radius of the particle. The results are compared with a limiting expression for the scattering from very elongated cylinders at very large angles, and it is shown that this limiting expression is a good approximation for all elongated cylinders over a wide angular range.

The subsidiary maxima are demonstrated experimentally in the scattering from solutions of randomly oriented tobacco mosaic virus.

Introduction

Information concerning the shape of colloidal particles may be obtained by examination of the angular intensity distribution of X-rays scattered at small angles from a colloidal solution. In practice, this is done by comparison of experimentally observed scattering distributions with those calculated from theoretical considerations for simple geometrical shapes.

The scattering is a maximum in the forward direction and drops off rapidly, as the angle increases, in a manner characteristic of the particle shape. To derive the scattering from a solution containing macromolecules of a particular shape; the scattering from a single particle of that shape is calculated as an average of all possible orientations of the figure with respect to the incident X-ray beam, and the scattering from the solution is proportional to this result. Thus, the following equations refer to solutions containing a single macromolecular specie, in which the molecules have neither preferred orientation nor spatial order. In addition, it must be noted that the results are rigorous only for molecules with a uniform electron distribution.

The theoretical intensity distribution has been derived for several simple geometrical figures. The simplest theoretical expression for the intensity distribution, that for a sphere, was originally derived by Rayleigh and is

$$i(h) = \Phi^2(ha) = \left[3 \frac{\sin ha - ha \cos ha}{h^3 a^3} \right]^2 \quad (1)$$

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where $h = 4\pi \sin \frac{1}{2}\theta/\lambda$; λ is the X-ray wavelength, θ is the scattering angle and a is the radius of the sphere. This, and the scattering functions following, are normalized for $i(0) = 1$.

The scattering from an ellipsoid of revolution was derived by Guinier (1939). For an ellipsoid of semi-axes a , a , va , his result may be written as

$$i(h) = \int_0^1 \Phi^2[ha\sqrt{1+y^2(v^2-1)}] dy, \quad (2)$$

where Φ^2 has the same functional dependence on its argument as in (1). Equation (2) cannot be integrated directly. Numerical solutions have been obtained for several axial ratios over an intensity range of $i(0)$ to about $10^{-2} \times i(0)$ by Guinier using a simple summation, and by Roess & Shull (1947) using hypergeometric series. Schmidt (1953) used Simpson's rule to evaluate the integral for slightly prolate ellipsoids ($v \leq 1.5$). Porod (1948) derived an equivalent power series expansion which he has calculated down to about $10^{-2} \times i(0)$.

Fournet (1951) has derived the scattering from a right circular cylinder of radius a and length $2va$; his result may be written as

$$i(h) = 4 \int_0^1 \frac{J_1^2(ha\sqrt{1-y^2}) \sin^2(havy)}{(ha\sqrt{1-y^2})^2 (havy)^2} dy, \quad (3)$$

where J_1 is the Bessel function of order 1. Porod has derived a power-series expansion for scattering from an elongated cylinder ($v > 1$) and evaluated it for several v 's to $10^{-2} \times i(0)$.

In the study of globular proteins by small-angle X-ray scattering, it is often possible to examine critically the scattering down to $10^{-4} \times i(0)$. With the exception of the scattering functions for spheres, which are well tabulated, and for the prolate ellipsoids cal-

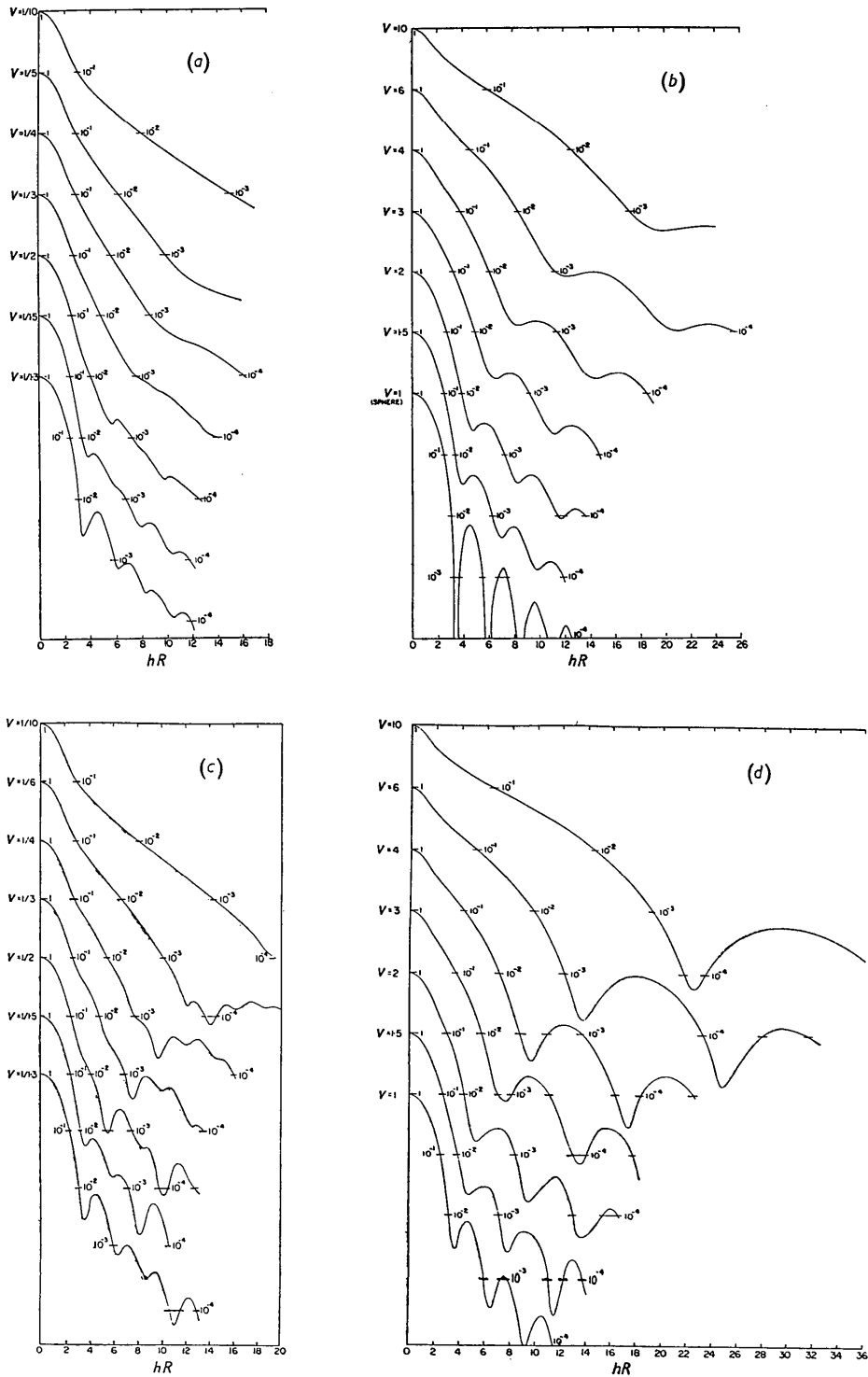


Fig. 1. (a) Scattering functions for oblate ellipsoids of revolution. (b) Scattering functions for prolate ellipsoids of revolution. (c) Scattering functions for flattened right circular cylinders. (d) Scattering functions for elongated right circular cylinders.

culated by Schmidt, intensity distributions are only known out to angles where the intensity is down to $10^{-2} \times i(0)$. In this range, the curves have no remarkable distinguishing features, and it is often difficult

to choose between several possible shapes. Thus it is desirable to have accurate calculations through a range at least as great as the experimental equipment is capable of detecting. Accordingly, the scattering

functions for ellipsoids of revolution and right circular cylinders were calculated with the aid of an electronic digital computer, the IBM Magnetic Drum Data-Processing Machine, Type 650, in the Numerical Analysis Laboratory of the University of Wisconsin.

Computation

Guinier has shown that the scattering curves of all particles with the same radius of gyration have the same limiting shape as $h \rightarrow 0$; therefore, the intensity was calculated (at half-integer intervals) as a function of hR where $R = [(2+v^2)/5]^{1/2}a$ is the radius of gyration for ellipsoids and $R = [(3+2v^2)/6]^{1/2}a$ is the radius of gyration for cylinders.

The equation for ellipsoids (2) was evaluated for axial ratios of $v = 1/10, 1/5, 1/4, 1/3, 1/2, 1/1.5, 1/1.4, 1/1.3, 1.5, 2, 3, 4, 6, 10$. That for cylinders (3) was evaluated for $v = 1/10, 1/6, 1/4, 1/3, 1/2, 1/1.5, 1/1.3, 1, 1.5, 2, 3, 4, 6, 10$. For most of these cases, the calculation includes an intensity range to $10^{-4} \times i(0)$.

The integrals were evaluated by Gauss's method for numerical quadrature. Each point was determined to an accuracy of at least 1%. The results are shown graphically* in Fig. 1; they agree (within corresponding regions) with those calculated by the authors mentioned above.

Discussion

Certain qualitative features of the scattering curves are apparent, and should be of value in choosing between elongated and flattened ellipsoids and cylinders. The elongated shapes show well defined, regularly spaced peaks of uniformly decreasing amplitude, in contrast with the flattened shapes, where the peaks, if present at all, are less pronounced, and irregular in spacing and amplitude.

The peaks for elongated cylinders are significantly more pronounced than those for the prolate ellipsoids. Figs. 1(b) and 1(d) show that, for elongated shapes, the position of the first peak is shifted to larger values of the abscissa as the axial ratio increases. The position of the first peak for prolate ellipsoids ($v \geq 1.5$) may be expressed empirically as $5.16[(2+v^2)/5]^{1/2}$ and for elongated cylinders ($v \geq 1$), $5.07[(3+2v^2)/6]^{1/2}$, with an accuracy of about 2%. That is, for both cases, the position of the first peak is a function of the equatorial radius of the particle. Thus if the elongated shapes were plotted against ha instead of hR , the first peaks would all occur at the same position along the abscissa, i.e., 5.16 for ellipsoids and 5.07 for cylinders. The situation is analogous for the second peaks; $ha = 8.54$ for ellipsoids and $ha = 8.21$ for cylinders. Several of the curves for cylinders include a third peak; for these, a common value of $ha = 11.5$ is found.

Porod (1948) has given a limiting expression for the scattering at large angles from very elongated cylinders of length $2H$; it is

* A limited quantity of these results in tabular form are available upon request.

$$i(h) = \frac{2\pi J_1^2(ha)}{v (ha)^3}, \quad v \geq 1, \quad hav \geq 1. \quad (4)$$

The first three maxima of (4) occur respectively at $ha = 5.04, 8.35, \text{ and } 11.6$. Thus it is seen that the peak positions given by this limiting expression for cylinders agree well for all cylinders of $v \geq 1$, and for prolate ellipsoids of $v \geq 1.5$.

In fact, (4) is a very good approximation to the scattering functions for cylinders of $v \geq 2$. This is demonstrated in Fig. 2, where points calculated from

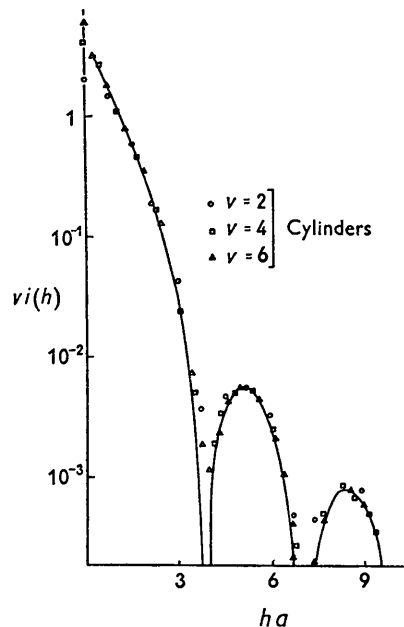


Fig. 2. Comparison of exact scattering functions for elongated ellipsoids with Porod's approximation (solid line).

the exact theoretical expression for several cylinders are superimposed on a curve representing Porod's approximation. This curve becomes infinite at $h = 0$ and extends to zero intensity at the minima, in contrast with the exact function which is finite in these regions. As would be expected, however, from the original limits imposed on (4), the differences become markedly less as v increases.

Application of results

The scattering functions presented here represent the scattering from a system satisfying the conditions mentioned above, i.e., random orientation, monodispersity, and no interaction between particles. Random orientation may be safely assumed in sufficiently dilute colloidal solutions. A polydisperse system will give a scattering curve which represents a weight average of the scattering from each component; thus, before an experimental curve is compared with the theoretical curves, independent examination of the solution is imperative to ensure that the solution contains a single macromolecular specie. As the average volume occupied by the particles approaches the

same order of magnitude as the total available volume, interparticle interference will result, and may become significant, especially at the smaller angles. This results in a distortion of the experimental curve; however, this may be minimized by using dilute solutions to obtain data in the very small angular range and more concentrated solutions for larger angles. In the very-small-angle region, the scattered intensity per particle is large; thus, a dilute solution may be used to minimize the interference effects which are important in this region. For larger angles, the scattered intensity per particle becomes small and the interference effects become insignificant; thus a concentrated solution may be used.

Finally, it should be noted that present experimental techniques do not render perfect collimation curves directly. Because the slits that collimate the incident and scattered X-rays are necessarily of finite width, an intensity measurement at a particular angular setting includes X-rays scattered over a range of angles about the nominal angle, and, for small angles especially, this introduces a significant distortion of the true intensity distribution. Thus for maximum utilization of the theoretical curves presented here, methods must be applied to reduce experimental curves to perfect collimation, or to 'smear' the perfect collimation curves to conform to the effects of a particular X-ray beam collimating system.

Scattering from tobacco mosaic virus

To demonstrate the subsidiary maxima noted above for elongated particles, the X-ray scattering from

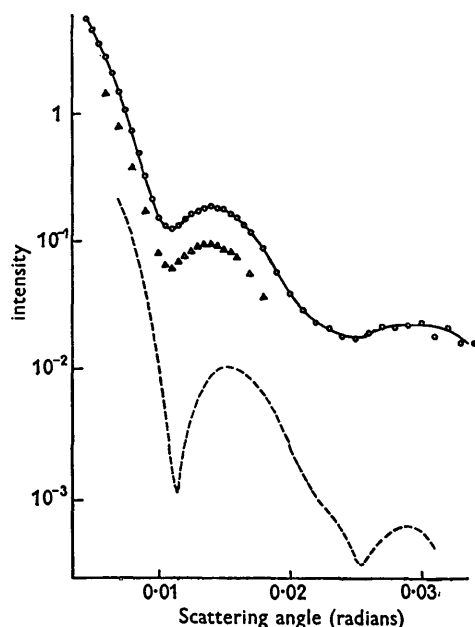


Fig. 3. Scattering curve from randomly oriented tobacco mosaic virus.

Circles: experimental points, 3% solution; triangles: experimental points, 1½% solution; dashed line: perfect collimation, 3% solution.

solutions of randomly oriented tobacco mosaic virus* was investigated.

The experimental results are shown in Fig. 3. The data for the 3% solution have been corrected for the smearing effects due to finite slit size, and the result is the lower (dashed) curve; this is the perfect collimation scattering curve, and thus may be compared with the results of the preceding sections.

Because it was not possible to get data at sufficiently small angles, $i(0)$ was not determined; consequently, the curves of Fig. 3 are not normalized.

Particular care was taken to ensure that the solutions under investigation had no oriented phase. It was examined for birefringence by placing it between cross polaroids while in the sample holder used for the scattering experiments; absence of birefringence, or tactoid formation, as described by Bernal & Fankuchen (1941), was taken as the criterion for random orientation. The agreement in shape of the scattering curves for the 3% and 1½% solutions gives further assurance that there was no preferred orientation which could affect the scattering, since preferred orientation is not associated with solutions of concentration less than 2%.

The expected subsidiary maxima are clearly observed. The first has its maximum at 0.0152 radians. According to the above results, this should be related to the cross-sectional radius, a , of the virus by $ha = 5.07$. Using this expression ($\lambda = 1.54 \text{ \AA}$), a is calculated to be 82 Å. This agrees well with the value of 84 Å measured by Caspar (1956).

For the second peak, the theoretical results predict $ha = 8.21$; the position of the second peak is 0.029 radians; from this, a is calculated to be 70 Å. The discrepancy between this and the radius calculated from the first peak is greater than experimental uncertainties; it can be explained by considering the internal structure of the virus since the theoretical results discussed here are rigorous only for particles of uniform electron distribution, and Caspar has shown that there are marked inhomogeneities in the electron distribution in the tobacco mosaic virus molecule.

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