Oosawa, F., Asakura, S., Hotta, K., Imai, N. & Ooi, J. (1959). *J. Polymer Sci.* **37**, 323–336.
Remenchik, A. P. & Bernsohn, J. (1967). *Anal. Biochem.* **18**, 1–9.
Shelanski, M. L., Gaskin, F. & Cantor, C. R. (1973). *Proc. Nat. Acad. Sci., U.S.A.* **70**, 765–768.
Tilney, L. G. & Porter, K. R. (1967). *J. Cell Biol.* **34**, 327–341.
Tilney, L. G., Hiramoto, Y. & Marsland, D. (1966). *J. Cell Biol.* **29**, 77–95.
Ventilla, M., Cantor, C. R. & Shelanski, M. L. (1972). *Biochemistry*, **11**, 1554–1561.
Weisenberg, R. C. (1972). *Science*, **177**, 1104–1105.

APPENDIX

Interpretation of the Light Scattering from Long Rods

BRUCE J. BERNE

Department of Chemistry
Columbia University
New York, N.Y. 10027, U.S.A.

When unpolarized incident light of wavelength λ is scattered from solutions containing macromolecules consisting of optically isotropic segments, the total intensity of the light scattered into the angle θ, regardless of the final polarization of the scattered light, is, in the Rayleigh–Gans limit (Landau & Lifshitz, 1960),

$$I(q) = cBk^4(1 + \cos^2\theta)S(q), \tag{A1}$$

where c is the number concentration of macromolecules, $k = 2\pi/\lambda$ is the wave number of the incident light, θ is the scattering angle, $S(q)$ is the macromolecular scattering factor about which we shall soon have more to say and B is a constant.

$$B = \frac{VE_0^2}{16\pi^2 R_0^2(\epsilon')^2}, \tag{A2}$$

where V is the scattering volume, E_0 is the amplitude of the incident light wave, R_0 is the distance of the detector from the scattering volume and ϵ' is the dielectric constant of the medium. B is thus seen to be a factor independent of angle, which can be essentially fixed in a sequence of measurements. It should be noted that k^4 gives rise to a λ^{-4} dependence of the intensity.

The quantity q that appears in equation (A1) is the magnitude of the Bragg vector:

$$q = (4\pi\mu/\lambda)\sin\tfrac{1}{2}\theta, \tag{A3}$$

where μ is the refractive index of the scattering medium. Thus in an unpolarized scattering experiment the scattered intensity depends on θ explicitly through the factor $(1 + \cos^2\theta)$ and implicitly through the dependence of $S(q)$ on q.

In the light-scattering experiments reported in the main text, the turbidity, h, is measured, where

$$h \equiv \frac{R_0^2}{V|E_0|^2}\int d\Omega I(q), \tag{A4}$$

where $\mathrm{d}\boldsymbol{\Omega}$ is an element of solid angle. This is proportional to the total intensity scattered into all scattering angles.

To evaluate h we must determine the scattering factor $S(q)$, which is by definition (Kerker, 1969):

$$S(q) = \langle | \sum_{j=1}^{N} \alpha_j \exp(\mathrm{i}\mathbf{q}\cdot\mathbf{r}_j)|^2 \rangle, \tag{A5}$$

where α_j and $\mathbf{r}_j$ are, respectively, the polarizability and position of the jth segment of the macromolecule and $\mathbf{q}$ is a vector whose magnitude is q and whose direction is $(\hat{\mathbf{k}} - \hat{\mathbf{k}}')$ where $\hat{\mathbf{k}}$ and $\hat{\mathbf{k}}'$ are, respectively, unit vectors pointing in the directions of the incident and scattered light.

In the subsequent discussion we adopt the following model of the microtubule:

(a) A tubule is a rigid rod consisting of optically isotropic segments of identical polarizability ($\alpha_j = \alpha_0$ for all j values) distributed uniformly along the rod axis.

(b) The thickness of a tubule is small compared to the wavelength of the incident light and compared with the length of the rod, L.

(c) The tubules are randomly oriented and the solutions are monodisperse.

As a consequence, $S(q)$ can be expressed as

$$S(q) = \alpha_0{}^2 S(q;\, L), \tag{A6}$$

where $S(q;\, L)$ is the well known structure factor of a uniform rigid rod of length L (Holtzer, 1955). For our purposes it is convenient to use a different explicit form of $S(q;L)$ than is usually used. This is

$$S(q;L) = L^2 \sigma(Q_0(1 - X)^{\frac{1}{2}}), \tag{A7}$$

where

$$\sigma(Q_0(1 - X)^{\frac{1}{2}}) \equiv \int_{-1}^{1} \mathrm{d}(\tfrac{1}{2}Y)|j_0(Q_0(1 - X)^{\frac{1}{2}}Y)|^2, \tag{A8}$$

and where

$$j_0(z) \equiv \sin z/z$$
$$Q_0 \equiv (2\pi\mu/\lambda)L/\sqrt{2}$$
$$X \equiv \cos\theta$$
$$Y \equiv (\mathbf{u}\cdot\mathbf{q})/q,$$

$\mathbf{u}$ being a unit vector specifying the rod axis. Q_0 depends on (L/λ) which is large in this case. X depends on the scattering angle θ, and Y is the cosine of the angle between the rod axis $\mathbf{u}$ and $\mathbf{q}$. The integral over Y arises from an average over a uniform distribution of rod orientations.

Substitution of equations (A1), (A6) and (A7) into equation (A4) gives

$$h_L = \left(\frac{c\alpha_0{}^2}{32\pi L^2(\epsilon')^2}\right) H(Q_0), \tag{A9}$$

where

$$H(Q_0) \equiv Q_0{}^4 \int_{-1}^{1} \mathrm{d}X(1 + X^2)\sigma(Q_0(1 - X)^{\frac{1}{2}}), \tag{A10}$$

where the subscript L indicates the dependence of h_L on L.

One important consequence of equation (A10) is that in the limit where the tubule is very long compared with λ, $Q_0 \gg 1$ and therefore in equation (A10) $H(Q_0)$ varies asymptotically as Q_0^3. This means that the turbidity is inversely proportional to λ^3 rather than to the usual λ^4 observed in Rayleigh scattering. In Figure (A1) we plot the apparent exponent,

$$m \equiv \left(\frac{\mathrm{d}\ln H(Q_0)}{\mathrm{d}\ln Q_0}\right), \tag{A11}$$

of $(1/\lambda)$ *versus* $\ln(Q_0{}^{-1})$. Note that large values of $\ln Q_0{}^{-1}$ correspond to wavelengths (λ) large compared to L, whereas negative values of $\ln Q_0{}^{-1}$ correspond to small λ values compared to L. In the former case $m \sim 4$, whereas in the latter case $m \sim 3$. Thus a measurement of h_L *versus* the wavelength λ defines the limit at which one is working, and may in fact be used to determine the approximate length of the rod.

Another important result is seen in Figure (A2), where $H(Q_0)/Q_0{}^2$ is plotted *versus* Q_0. For sufficiently large Q_0 values this function is a linear function of Q_0, or better yet L. Since h_L is directly proportional to $cH(Q_0)/Q_0{}^2$, with a proportionality constant independent of L, it follows that the turbidity is directly proportional to cL, or

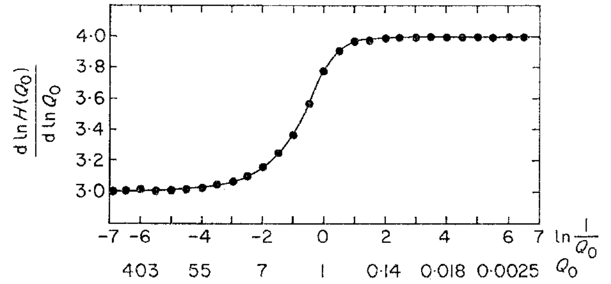

FIG. A1 The relation between the apparent exponent eqn (A11) $m \equiv (\mathrm{d}\ln H(Q_0)/\mathrm{d}\ln Q_0)$ of $1/\lambda$, as a function of $\ln(Q_0{}^{-1})$.

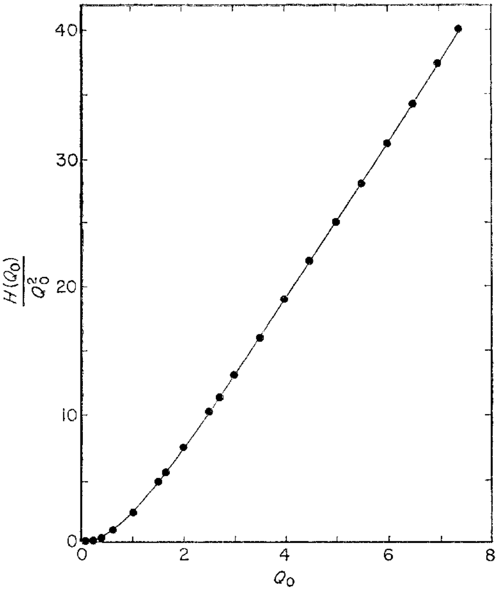

FIG. A2. The variation of $H(Q_0)/Q_0{}^2$ *versus* Q_0, where $H(Q_0)/Q_0{}^2$ is defined in eqn (A10).

concomitantly to the total number, N, of monomeric units in the tubule solution, i.e.

$$h \propto N$$

This is the result which is crucial to the analysis described in the main text.

The macromolecular solutions are rarely monodisperse. In the case of polydispersity we must average h_L over a distribution of lengths. This does not change any of the above conclusions provided that the macromolecules present are sufficiently long compared to λ.

In conclusion, we should like to emphasize that these calculations were carried out in the Rayleigh–Gans limit and are restricted to the requirements of that limit.

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REFERENCES

Holtzer, A. M. (1955). *J. Polymer Sci.* **17**, 432–434.

Kerker, M. (1969). *The Scattering of Light and Other Electromagnetic Radiation*, Academic Press, New York.

Landau, L. D. & Lifshitz, E. M. (1960). *Electrodynamics of Continuous Media*, Addison Wesley Publishing Co., Inc., Reading, Mass.