PHYSICAL CHEMISTRY

B. A. FEDOROV, O. B. PTITSYN

1963

SovietRxiv

Fig. 1. Curves of the dependence of $P_{\mathrm{pl}}(\theta)\gamma^2$ and $P_{\mathrm{cyl}}(\theta)\gamma^2$ on γ ($a = c$ and $b/c = 100$)

Figure 1: Fig. 1. Curves of the dependence of $P_{\mathrm{pl}}(\theta)\gamma^2$ and $P_{\mathrm{cyl}}(\theta)\gamma^2$ on γ ($a = c$ and $b/c = 100$)

Abstract

Full Text

PHYSICAL CHEMISTRY

B. A. FEDOROV, O. B. PTITSYN

ON THE DETERMINATION OF THE TRANSVERSE DIMENSIONS OF MACROMOLECULES BY THE METHOD OF SMALL-ANGLE X-RAY SCATTERING

(Presented by Academician V. A. Kargin, June 29, 1963)

The study of small-angle X-ray scattering, as is known ($^{1-5}$), provides a number of important data on the structure of molecules of polymers and biopolymers in solution, in particular on the distribution of scattering centers over short distances. In particular, for rodlike particles, from the slope of the curve of the dependence $\ln[i(\theta)\sin\theta] \simeq \ln[i(\theta)\theta]$ on $\sin^2\theta \simeq \theta^2$ ($i(\theta)$ is the scattering intensity at angle 2θ), one can determine the radius of inertia of the transverse section of the macromolecule ($^{6-8}$), which has been used to determine the diameter of DNA (^{9,10}) and RNA (^{4}) helices.

Fig. 1. Curves of the dependence of $P_{\mathrm{pl}}(\theta)\gamma^2$ and $P_{\mathrm{cyl}}(\theta)\gamma^2$ on γ ($a = c$ and $b/c = 100$)

In the present communication a more sensitive method will be proposed for determining the transverse dimensions of macromolecules from the curve of the dependence of $i(\theta)\theta^2$ on θ. As is known (^{11,12}), this curve has a bend, the position of which makes it possible to determine the persistent length of the chain (i.e., its flexibility). We shall show that at large θ the indicated curve passes through a maximum, the position of which is determined by the transverse dimensions of the chain. Indeed, the relative scattering intensity by a circular cylinder of height b and diameter a, under the condition $k \equiv b/a \gg 1$, has the form (^{7})

$$P_{\mathrm{cyl}} \equiv \frac{i(\theta)}{i(0)} = \frac{16\pi}{k}\frac{J_1^2(\gamma/2)}{\gamma^3}, \tag{1}$$

where $\gamma = \frac{4\pi a}{\lambda} \sin\theta$ (λ is the wavelength), and $J_\nu(\gamma)$ is a Bessel function of order ν. Calculation shows that the function $P_{\text{cyl}}(\theta)\gamma^2$ as a function of γ passes through a maximum (see Fig. 1), the position of which is determined by the equation $\gamma J_0(\gamma/2) = 3J_1(\gamma/2)$, with solution $\gamma_1 = 2.72$.

Formula (1) and the expression for γ_1 based on it are valid when a point source of X-rays is used. In practice, sources with slit collimation are usually used, which must introduce a certain correction. If $i(s)$ is the X-ray scattering intensity from a point source ($s = 2\sin\theta/\lambda$), and $j(s)$ is that from a slit source, then (^{1})

$$j(s) = \int_{-\infty}^{\infty} i\left(\sqrt{s^2 + t^2}\right) dt, \tag{2}$$

To obtain the function $j(s)$ in a convenient form, Luzzati (^{8}) adopted, instead of (1),

$$i(s) \sim \frac{1}{s} \exp(-2B^2 s^2),$$

where $B = \pi a / 2\sqrt{2}$, which gives

$$j_1(s) \sim \int_{-\infty}^{\infty} \frac{e^{-2B^2(s^2+t^2)}}{(s^2+t^2)^{1/2}}\, dt \sim e^{-B^2 s^2} K_0(B^2 s^2), \tag{3}$$

where

$$K_\nu(x) = \frac{\pi i}{2} e^{\pi \nu i/2} H_\nu^{(1)}(x)$$

($H_\nu^{(1)}(x)$ is a Hankel function).

The analysis we have carried out of formula (3) shows that the function $j_1(s)s^2$ passes through a maximum whose abscissa is determined by the equation

$$K_0(x) = \frac{x}{1-x} K_1(x) \quad (x = B^2 s^2)$$

with the solution $B^2 s^2 = 0.32$, or $\gamma_2 = 3.20$.

To obtain the abscissa of the maximum for slit collimation, using the exact formula (1), the integral * was calculated on the BESM-2 electronic computer of the Computing Center of the Leningrad Branch of the V. A. Steklov Mathematical Institute of the Academy of Sciences of the USSR:

$$j_2(s)s^2 \sim \int_0^{\infty} \frac{J_1^2\left(2\sqrt{2}\, B \sqrt{s^2+t^2}\right)}{(s^2+t^2)^{3/2}}\, dt \sim \int_0^{\pi/2} J_1^2\left(\frac{\gamma}{2\cos\varphi}\right) \cos\varphi\, d\varphi, \tag{4}$$

which gives a maximum at $\gamma_3 = 2.92$. Thus, the abscissae of the maxima with and without allowing for the slit-collimation correction (γ_1 and γ_3) are very close, which makes it possible, in a first approximation, not to take the collimation correction into account (see below).

If the transverse dimensions of the macromolecule in one direction are appreciably greater than in the other, then it may be modeled not by a cylinder but by a narrow (two-dimensional) plate. For a plate of length b and width c we have

$$P_{\mathrm{pl}}(\vartheta) = \int_0^1 \int_0^1 \int_0^1 \int_0^1 \frac{\sin\left[\gamma^* \sqrt{(x-x')^2 + \chi^2 (y-y')^2}\right]}{\gamma^* \sqrt{(x-x')^2 + \chi^2 (y-y')^2}}\, dx\, dx'\, dy\, dy', \qquad (5)$$

where $\gamma^* = 2\pi cs$, $\chi = b/c$. Since, obviously,

$$\int_0^1 \int_0^1 \int_0^1 \int_0^1 f[(x-x'), (y-y')]\, dx\, dx'\, dy\, dy' = 4 \int_0^1 dt \int_0^1 f(t, t')(1-t)(1-t')\, dt', \qquad (6)$$

the problem reduces to considering the integrals

$$L_1 = \int_0^1 \int_0^1 f(t, t')\, dt\, dt', \qquad L_2 = \int_0^1 \int_0^1 t f(t, t')\, dt\, dt',$$

$$L_3 = \int_0^1 \int_0^1 t'(1-t) f(t, t')\, dt\, dt',$$

where

$$f(t, t') = \frac{\sin\left[\gamma^* \sqrt{t^2 + \chi^2 t'^2}\right]}{\gamma^* \sqrt{t^2 + \chi^2 t'^2}}.$$

* The calculations were performed by A. K. Kron, to whom the authors express their sincere gratitude.

We have:

$$L_1 = \frac{1}{\gamma^* \chi} \int_0^1 dt \left[\int_{\gamma^* t}^{\infty} \frac{\sin x\, dx}{\sqrt{x^2 - \gamma^{*2} t^2}} - \int_{\gamma^* \sqrt{t^2 + \chi^2}}^{\infty} \frac{\sin x\, dx}{\sqrt{x^2 - \gamma^{*2} t^2}} \right] = \frac{\pi}{2\gamma^* \chi} \int_0^1 J_0(\gamma^* t)\, dt$$
$$= \frac{\pi}{2\gamma^{*2} \chi} \left\{ \gamma^* J_0(\gamma^*) + \frac{\pi \gamma^*}{2} \left[J_1(\gamma^*) H_0(\gamma^*) - J_0(\gamma^*) H_1(\gamma^*) \right] \right\}. \qquad (7)$$

Fig. 2. Experimental points from work (^{9}), replotted in the coordinates $\log[P\gamma^2]$ and $\log\gamma$ (it is conventionally assumed that $a = 120$ Å)

Figure 2: Fig. 2. Experimental points from work (^{9}), replotted in the coordinates $\log[P\gamma^2]$ and $\log\gamma$ (it is conventionally assumed that $a = 120$ Å)

($J_\nu(\gamma^*)$ and $H_\nu(\gamma^*)$ are, respectively, Bessel and Struve functions), since for $c^2/b^2 \ll$

$$\int_{\gamma^*\sqrt{t^2+\chi^2}}^{\infty} \frac{\sin x\,dx}{\sqrt{x^2-\gamma^{*2}t^2}} \ll \frac{1}{\gamma^*\chi},$$

i.e., in the region where $\gamma^* \gg 1$ (i.e., $\gamma^*\chi \gg 1$), this integral may be neglected;

$$L_2 = \frac{1}{\gamma^*\chi}\int_0^1 t\,dt \int_{\gamma^* t}^{\gamma^*\sqrt{t^2+\chi^2}} \frac{\sin x\,dx}{\sqrt{x^2-\gamma^{*2}t^2}} = \frac{\pi}{2\gamma^*\chi}\int_0^1 tJ_0(\gamma^* t)\,dt = \frac{\pi}{2\gamma^*\chi}J_1(\gamma^*), \tag{8}$$

$$L_3 \sim \frac{1}{\gamma^{*2}\chi^2} \ll L_1, L_2,$$

therefore, for $\gamma^* > 1$,

$$P_{\text{pl}}(\theta) = \frac{\pi}{\gamma^*\chi}\left\{2\left[J_0(\gamma^*) - \frac{J_1(\gamma^*)}{\gamma^*}\right] + \pi\left[J_1(\gamma^*)H_0(\gamma^*) - J_0(\gamma^*)H_1(\gamma^*)\right]\right\}. \tag{9}$$

When $c = 0$, the expression in braces becomes unity, and formula (9) (as does (1) when $a = 0$) becomes the formula for scattering by a rod in the region $2\pi sb \gg 1$ (see, for example, (^{1})). The curve of the dependence of $P_{\text{pl}}(\theta)\gamma^{*2}$ on γ^* (see Fig. 1) passes through a maximum and then begins to oscillate, tending to a constant value; moreover, as can be seen from an analysis of formula (9), the abscissas of the extrema of the function $P_{\text{pl}}(\theta)\gamma^{*2}$ are the roots of the equation $J_1(\gamma^*) = 0$. The first maximum corresponds to $\gamma^* = 3.83$. Thus, the behavior of the curves $P_{\text{pl}}(\theta)\gamma^2$ to the right of the maximum makes it possible to choose between the models under consideration, and after the model has been chosen, to determine quantitatively the transverse dimensions of the macromolecule.

Fig. 2. Experimental points from work (^{9}), replotted in the coordinates $\log[P\gamma^2]$ and $\log\gamma$ (it is conventionally assumed that $a = 120$ Å).

Most macromolecules are modeled better by a cylinder than by a narrow plate. The experimental data for these macromolecules lead to the above-described picture of X-ray scattering by a cylinder, and the values obtained for the thickness of the molecule are close to the values calculated from the known conformations of these chains. As has already been reported (5), our replotting of the experimental X-ray scattering curves for molecules of various samples of ribosomal RNA (4) in the coordinates $i(\theta)\theta^2 - \theta$ gives curves with clearly expressed maxima, whose abscissas lead to a helix diameter of ~ 22 Å, in good agreement with the diameter of the Crick–Watson helix. Figure 2 gives an analogous treatment of the experimental X-ray scattering curves for native DNA molecules obtained in work (9). The position of the maximum in this case corresponds to a helix diameter of ~ 22 Å.

Finally, the curves of the dependence of $i(\theta)\theta^2$ on θ for polymethyl methacrylate in various solvents (13) have maxima at $\theta = 3°50'$, whose nature remained unexplained in work (13). In light of what has been set forth above

It is obvious that these maxima are due to scattering by the transverse dimensions of the macromolecule and correspond to a helix diameter of 5.25 ± 0.05 Å, i.e., an entirely reasonable value.

As for the sheet-like model, the proposed theory may prove useful for interpreting X-ray scattering curves, for example, by amylose macromolecules, in which the glycosidic rings lie practically in one plane, so that the "thickness" of the chain is considerably less than its "width."

Leningrad State University
named after A. A. Zhdanov

Institute of High-Molecular Compounds
Academy of Sciences of the USSR

Received
29 VI 1963

REFERENCES

1. A. Guinier, G. Fournet, *Small-angle Scattering of X-rays*, N. Y., 1955.
2. O. Kratky, *Makromolek. Chem.*, **35a**, 12 (1960).
3. O. Kratky, *Koll. Zs.*, **182**, 7 (1962).
4. S. N. Pimasheff, J. Witz, V. Luzzati, *Biophys. J.*, **1**, 525 (1961).
5. O. B. Ptitsyn, B. A. Fedorov, *Tsitologiya*, **5**, 355 (1963).
6. O. Kratky, G. Porod, *Acta Phys. Austr.*, **2**, 133 (1948).
7. G. Porod, *Acta Phys. Austr.*, **2**, 255 (1948).

8. V. Luzzati, *Acta Crystallogr.*, **13**, 939 (1960).

9. V. Luzzati, A. N. Nicolaeff, F. Masson, *J. Mol. Biol.*, **3**, 185 (1961).

10. V. Luzzati, D. Luzzati, F. Masson, *J. Mol. Biol.*, **5**, 375 (1962).

11. O. Kratky, G. Porod, *Rec. Trav. chim.*, **68**, 1106 (1949).

12. B. A. Fedorov, T. M. Birshtein, O. B. Ptitsyn, *Biofizika*, **8**, 288 (1963).

13. R. Kirste, O. Kratky, *Zs. phys. Chem.*, Neue Folge, **31**, 363 (1962).

Note: Figure translations are in progress. See original paper for figures.

Source: Math-Net.Ru and CyberLeninka. Machine translation. Verify with the original.