Overcoming Angular Dependency When Teaching Light Scattering Using a Spectrofluorometer: The Molecular Weights of Latex Beads

W

Nuno C. Santos[†]

Centro de Química Física Molecular, Instituto Superior Técnico, Lisboa, Portugal

Miguel X. J. J. Fernandes

Departamento de Química e Bioquímica, Faculdade de Ciências da Universidade de Lisboa, Campo Grande, Lisboa, Portugal

Miguel A. R. B. Castanho[*†]

Centro de Química Física Molecular, Complexo I, Instituto Superior Técnico, 1049-001 Lisboa, Portugal; *pcmcastanho@alfa.ist.utl.pt

The practical teaching of light scattering technique has been the focus of a number of papers (*1–4*). Some of them are dedicated only to static (also called "total intensity") light scattering, and spectrofluorometers or UV–vis absorption spectrophotometers are used instead of specialized equipment. Spectrofluorometers are ubiquitous in teaching laboratories, easy to use and fairly inexpensive. However, if a spectrofluorometer is used in these experiments, it is impossible to carry out angle-dependent studies. At first glance, this seems to students to be an insurmountable obstacle. Nevertheless, a spectrofluorometer presents potentialities not available in classical laser light scattering spectroscopy, namely, the availability of much broader wavelength ranges. Changing the wavelength changes the magnitude of the scattering vector, leading to the same effect as changing the measurement angle:

$$q = \frac{4\pi n_0}{\lambda} \sin\left(\frac{\theta}{2}\right) \qquad (1)$$

(q is the magnitude of the scattering vector, n_0 is the refractive index of the medium, λ is the wavelength in vacuum and θ is the measurement angle). Thus, it is possible to overcome angle dependency by accounting for wavelength dependency. This "shift" in dependencies is easily understood by students because it results directly from the usual formalisms used in light scattering. Moreover, this approach encourages students, in practice, to elaborate innovative solutions, even from classical formalisms. This methodology is applied to the calculation of a series of molecular weights of polystyrene latex spheres. This material has several advantages: (i) water miscibility, (ii) definite shape, (iii) monodispersity (both size and shape), (iv) low cost, and (v) availability in a wide variety of sizes and composition. Although they have been very well characterized with respect to size and shape (mainly by dynamic light scattering and electronic microscopy studies), latex spheres have been poorly studied in terms of molecular weight. Working in an innovative area of research proves stimulating for students. This work is a challenge for graduate and experienced undergraduate students working in physical chemistry and physical biochemistry, in the sense that it allows them to appreciate how apparent experimental limitations (in this case an apparent instrumental limitation) can be overcome and how knowledge of even an intensively studied system can be enlarged experimentally. Moreover, this work forces the students to deal with basic concepts that underlie the function of a spectrofluorometer (e.g., the dependence of monochromator performance on wavelength and photomultiplier sensitivity).

The results obtained from application of this new methodology were compared to results obtained from classical light scattering (Zimm plot). A reasonable agreement is obtained for the weight average molecular weight, but the agreement in the second virial coefficient is only qualitative. In the analysis, the concept of intra-particle structure factor and the relationship between the molecular weight as calculated by light-scattering techniques and density-based techniques are simply and clearly dealt with.

Although the methodology described here was tested and illustrated by using latex beads, for the reasons mentioned above, it can be applied to other systems, such as globular proteins and the tobacco mosaic virus.

Acknowledgments

Note

[W]Supplementary materials for this article are available on *JCE Online* at *http://jchemed.chem.wisc.edu/Journal/issues/1999/Sep/abs1259.html*. The document includes notes to the instructor, presentation of the theoretical background, details of the method and data treatment, a discussion, and additional references to the literature.

Literature Cited

1. Thompson, A. C.; Kozimer, K. G.; Stockwell, D. *J. Chem. Educ.* **1970**, *47*, 828–831.
2. Mathews, G. P. *J. Chem. Educ.* **1984**, *61*, 552–554.
3. Mougán, M. A.; Coello, A.; Meijide, F.; Vato, J. V. *J. Chem. Educ.* **1995**, *72*, 284–286.
4. Santos, N. C.; Castanho, M. A. R. B. *Biophys. J.* **1996**, *71*, 1641–1650.

[†]Also: Departamento de Química e Bioquímica, Faculdade de Ciências da Universidade de Lisboa, Campo Grande, Lisboa, Portugal.