SAXSLAB

Absolute intensity calibration with glassy carbon

Introduction

While for many applications it is not necessary to have the scattering data displayed on absolute scale, it does provide a useful tool to assess data quality, and it is essential for the comparison of data from different instruments, to obtain molecular weights and to determine the volume fraction of different species.[1–7] When the measured intensity is corrected for the intensity of the incoming beam, the measurement time, transmission of the sample, the volume the beam passes trough, and the solid angle, it only depends on the scattering cross-section of the material.

There are two intrinsically different methods to obtain scattering curves on an absolute scale. Primary calibration, based on the geometry and intensity of the beam of the used instrument,[8] and secondary calibration, based on the use of a known standard. This standard can either be a primary standard of which the scattering properties can be calculated – most commonly water,[6] but any other pure solvent can be used.[1] Or a secondary standard, which is a previously calibrated standard, such as a protein solution of a known concentration[5] or glassy carbon.[7]

Glassy carbon

Glassy carbon, sometimes called vitreous carbon, is a form of carbon of which the atomic structure has not been elucidated yet. It is often used for high temperature applications and as electrode material. The glassy carbon that is widely used for absolute intensity scaling consists of plates of 50x50x10 mm obtained from Alfa Aesar, which are calibrated at the Advanced Photon Source at Argonne National Laboratory.[9]

It is a great standard for the calibration of lab instruments because of its high scattering intensity in the range between 0.1 and 1 $Å^{-1}$ (Fig 1). In contrast to the use of water, which due to its low scattering cross-section requires long measurements to obtain good statistics, the measurements of glassy carbon are relatively short. Moreover, since the scattering is so strong, no background measurement is required, which further shortens the calibration routine.

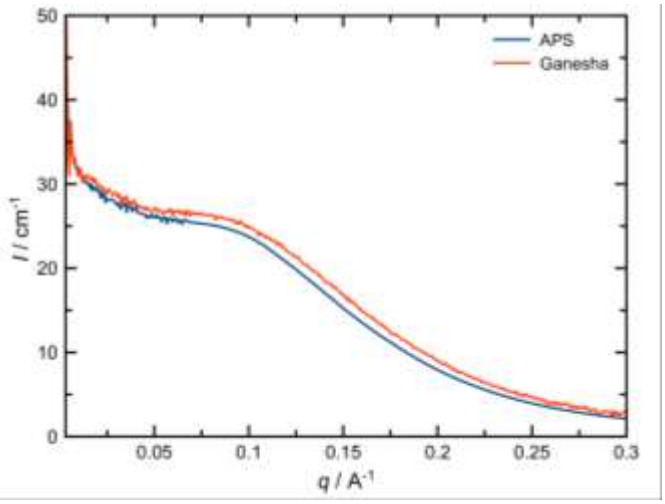

Fig 1. Radially averaged scattering pattern of glassy carbon (sample M12).

Calibration

To convert your data to absolute intensity using Glassy Carbon as a secondary standard you need to determine the scaling factor for your instrument configuration. This is done by measuring the glassy carbon data and scaling it to the calibration curve that belongs to that particular piece of glassy carbon, specifically the plateau at q = 0.006Å^{-1}.

Figure 1, shows such a comparison, resulting in a calibration factor of 0.95. The data is actually a comparison of the data from 2 systems, which each try obtain the correct absolute scattering intensity by primary standards: The APS system and the GANESHA. The results lie within 5% of each other, which is certainly within experimental uncertainty. For a full description of the procedure and an instructional YouTube video, have a look at ref. 9.

Concluding remarks

When available, calibrated glassy carbon is freely obtainable from Jan Ilavsky.[9] It is advisable to check the thickness of the sample.

References and further reading

1. Dreiss, C. A., Jack, K. S. & Parker, A. P. On the absolute calibration of bench-top small-angle X-ray scattering instruments: a comparison of different standard methods. *J. Appl. Crystallogr.* **39,** 32–38 (2006).
2. Pauw, B. R. Everything SAXS: small-angle scattering pattern collection and correction. *J. Phys. Condens. Matter* **25,** 383201 (2013).
3. Jacques, D. A. & Trewhella, J. Small-angle scattering for structural biology—Expanding the frontier while avoiding the pitfalls. *Protein Sci. Publ. Protein Soc.* **19,** 642–657 (2010).
4. Wignall, G. D. & Bates, F. S. Absolute calibration of small-angle neutron scattering data. *J. Appl. Crystallogr.* **20,** 28–40 (1987).
5. Mylonas, E. & Svergun, D. I. Accuracy of molecular mass determination of proteins in solution by small-angle X-ray scattering. *J. Appl. Crystallogr.* **40,** 245–249 (2007).
6. Orthaber, D., Bergmann, A. & Glatter, O. SAXS experiments on absolute scale with Kratky systems using water as a secondary standard. *J. Appl. Crystallogr.* **33,** 218–225 (2000).
7. Zhang, F. *et al.* Glassy Carbon as an Absolute Intensity Calibration Standard for Small-Angle Scattering. *Metall. Mater. Trans. A* **41,** 1151–1158 (2009).
8. Zemb, T., Tache, O., Né, F. & Spalla, O. A high sensitivity pinhole camera for soft condensed matter. *J. Appl. Crystallogr.* **36,** 800–805 (2003).
9. http://usaxs.xray.aps.anl.gov/docs/glassy_carbon_standard/