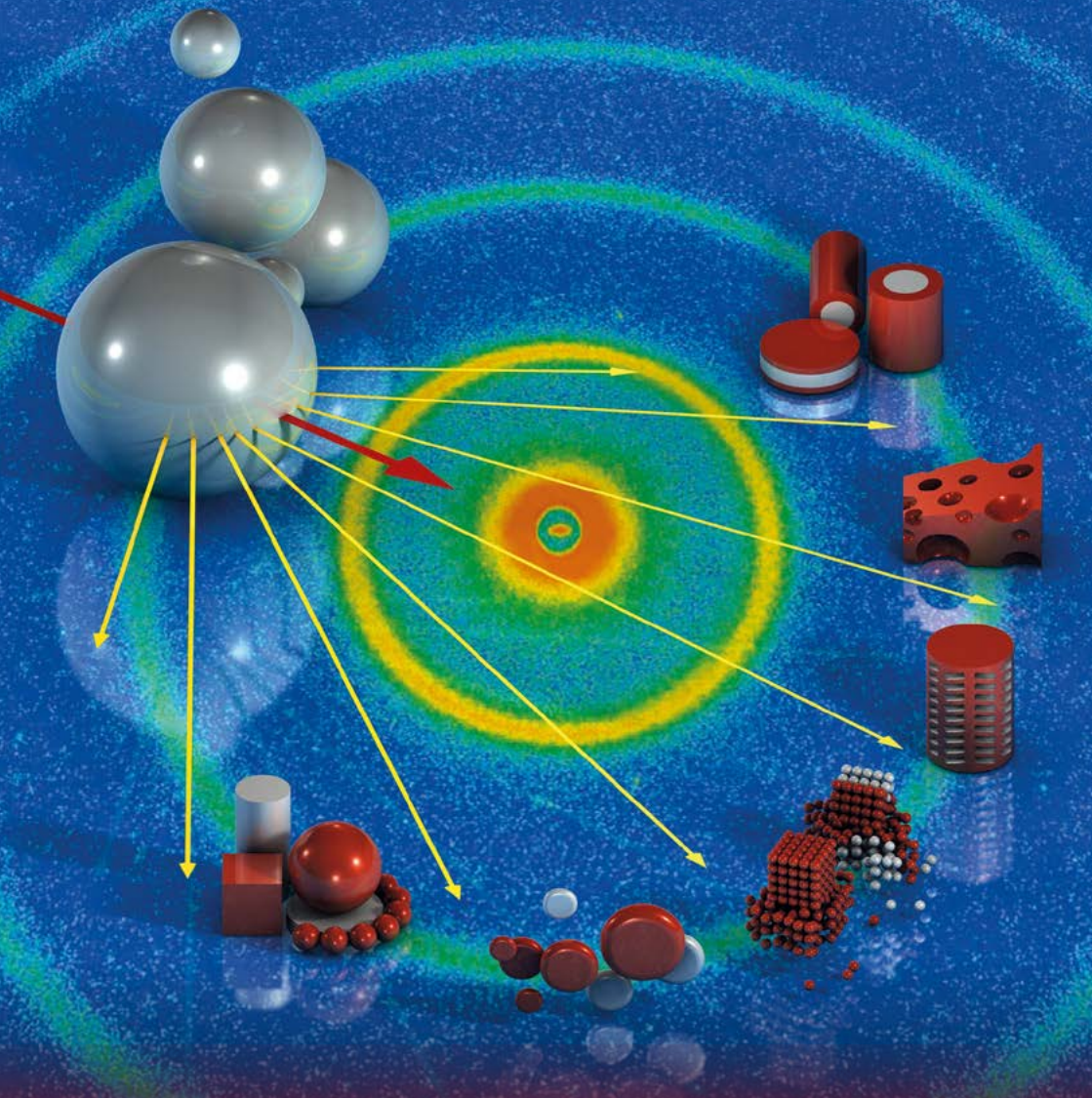
The SAXS Guide

Getting acquainted with the principles



New Edition with special contributions

The SAXS Guide

Getting acquainted with the principles

4th edition

by
Heimo Schnablegger
Yashveer Singh

Special contributions on

"SAXS Structural Biology" by Jill Trewhella,
The University of Sydney and Tobias Madl,
Medical University of Graz

and

"GISAXS – Grazing-Incidence Small-Angle Scattering"
by Detlef-M. Smilgies, Cornell Biomolecular Synchrotron
Source (CHESS)

➤ Published by Anton Paar GmbH. Printed in Austria.

Anton Paar GmbH
Anton-Paar-Str. 20
A-8054 Graz
Austria - Europe
Tel.: +43 316 257-0
Fax: +43 316 257-257
E-Mail: info@anton-paar.com
Web: www.anton-paar.com

Date: June 2017

Specifications subject to change without notice. | 06/17 D21IP004EN-B

Contents

1. Introduction

This document gives a general introduction to Small-Angle X-ray Scattering (SAXS) and SAXS analysis. It explains how a SAXS instrument works and how SAXS analysis is done. It is intended to help people new to the field of SAXS analysis. Difficult mathematical equations are avoided and the document requires only basic knowledge of mathematics, physics and colloid chemistry. The advanced reader is also encouraged to look for details in the original literature[1]-[6], which can be found in the references section (see „Literature“ on page 144).

This document is not dedicated to one specific scattering instrument or one particular application area, but aims to give a global overview of the main instrumentation and applications.

2. What is SAXS

SAXS is an analytical method to determine the structure of particle systems in terms of averaged particle sizes or shapes. The materials can be solid or liquid and they can contain solid, liquid or gaseous domains (so-called particles) of the same or another material in any combination. Normally, X-rays are sent through the sample (transmission mode) and every particle that happens to be inside the beam will send out its signal. Thus, the average structure of all illuminated particles in the bulk material is measured.

But also surface-near particles can be measured selectively, when the X-rays hit a flat sample almost parallel to its surface and the scattering signal is measured in reflection mode. This discipline of SAXS is called GISAXS (GI = grazing incidence) and it measures the average structure of all illuminated particles and their relative positional order on the surface or within the surface layer.

The SAXS method is accurate, non-destructive and usually requires only a minimum of sample preparation. Application areas are very broad and include biological materials, polymers, colloids, chemicals, nanocomposites, metals, minerals, food and parmaceuticals and can be found in research as well as in quality control.

The samples that can be analyzed and the time requirements of the experiments mainly depend on the used instrumentation, which can be classified into two main groups, (1) the line collimation instruments and (2) the point collimation instruments, which are explained in more detail later. The particle or structure sizes that can be resolved range from 1 to 100 nm in a typical set-up but can be extended on both sides by measuring at smaller (Ultra Small-Angle X-Ray Scattering, USAXS) or larger angles (Wide-Angle X-Ray Scattering, WAXS also called X-Ray Diffraction, XRD) than the typical 0.1° to 10° of SAXS. The concentration ranges between 0.1 wt.% and 99.9 wt.%. Generally speaking, particles made of materials with high atomic numbers show higher contrast and have lower detection limits when measured in matrix materials

of lighter elements. Matrix materials of heavy elements should be avoided due to their high absorption of X-rays.

Standards are required only in the following two situations:

1. When the **sample-to-detector distance** is not known. Then a reference sample of known structure is measured in order to calibrate the scattering angles. This is required only for instruments that employ unreliable mechanical movements and have poorly documented detector or sample positions.

2. When the **number density** of particles or their **mean molecular weight** has to be determined. Then the experimental intensities must be scaled by the intensity from a standard sample, such as water. For the determination of the particle structure, however, this is not required at all.

Fig. 2.-1 shows a typical pair of scattering profiles of a dispersion of particles and of the solvent alone. The difference between these two profiles is the actual signal and is put into calculations in order to obtain the information of size, shape, inner structure or the specific surface of the particles.

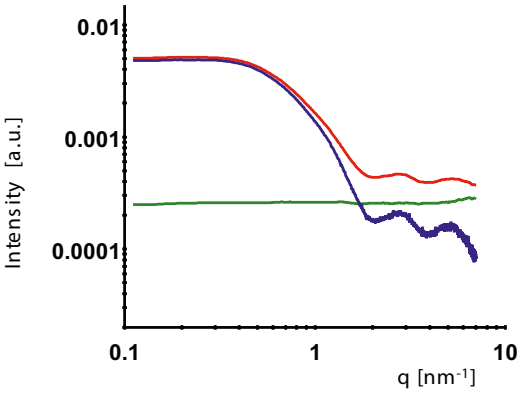


Fig. 2.-1.
Typical SAXS profiles of (red) a particle dispersion, (green) of the solvent and (blue) the difference profile therefrom.

➤ 2.1. Scattering and microscopy

Scattering and absorption are the first processes in any technique that uses radiation, such as an optical microscope (see Fig. 2.-2). This means that interaction between matter and the incoming radiation must take place. Otherwise no picture of the investigated object (= particle) will be available. Neither with microscopy nor with scattering can an object be investigated, when there is no contrast. In order to establish contrast in SAXS, the particles must have an electron density different than that of the surrounding matrix material (e.g., the solvent).

Although the operation of a scattering instrument is identical to the first process that takes place in a microscope, its result is complementary to that of a microscope, as will be outlined below.

The second process in an optical microscope is the reconstruction of the object (particle) from the scattering pattern (see Fig. 2.-2). This is done with the help of a lens system. If a lens system is not readily available for the used radiation (such as X-rays), then a reconstruction is not directly possible. Instead, the scattering pattern must be recorded and the reconstruction must be attempted in a mathematical way rather than in an optical way.

In the recording process the phases of the detected waves are lost. This constitutes the main difference between microscopy and X-ray scattering. Because of the lost phases, it is not possible to achieve a 3D (holographic) representation of the object in a direct way, as it would be possible with a lens system.

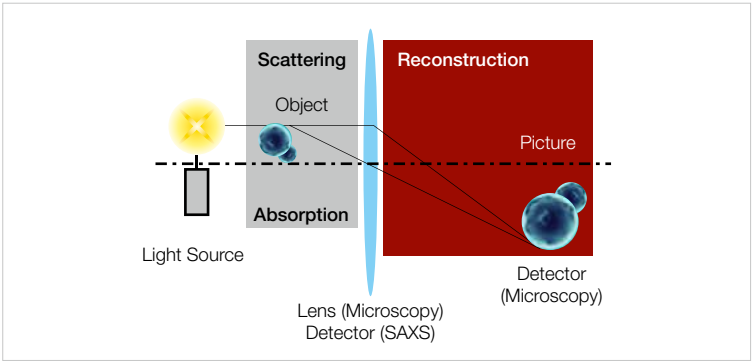


Fig. 2.-2.
The first processes of a microscopic investigation are scattering and absorption. Microscopy: The scattered waves are processed into a picture (reconstructed) by a lens. SAXS: The scattered intensity is recorded by a detector and is processed mathematically, as a replacement for the actions of a lens.

In microscopy one object or a small part of a sample is magnified and investigated. With scattering techniques the whole illuminated sample volume is investigated. As a consequence, average values of the structure parameters are obtained by SAXS. The average is taken over all objects and over all orientations of the objects. Therefore, structure details of the object will not become visible unless they are pronounced enough in the whole sample and are therefore representative.

The signal strength in SAXS scales with the squared volume of the particle. This means that small particles are hardly visible in the presence of big particles. On the other hand, SAXS is very sensitive to the formation or growth of large particles.
The resolution criteria in SAXS are the same as those in microscopy. The closer the lens to the object (the larger the aperture or the scattering angle), the smaller is the detail that can be resolved. The farther away the object is from the lens (the smaller the aperture or the scattering angle), the bigger is the largest object that can be brought into the picture.

The following table summarizes a typical comparison of the two techniques.

Feature	Microscopy	Scattering
Small details are	visible	not visible
Results are	unique but not representative	representative but ambiguous
Local structure details	can be extracted	cannot be extracted
Average structures are	hard to obtain	always obtained
Preparation artifacts are	inherent	scarce (in vitro experiments)

In order to get the complete picture of an unknown sample one needs to make use of both methods, because **their results are complementary.**

Thank you for your interest in the SAXS Guide.

To receive a **free hard copy** of the guide, please **fill out the request form**.

➤ www.anton-paar.com/saxs-guide/

Enjoy reading!