

The Small Angle X-ray Scattering Technique: An Overview

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An Overview

X-ray scattering techniques are a family of non-destructive analytical techniques which reveal information about the crystallographic structure, chemical composition, and physical properties of materials and thin films. These techniques are based on observing the scattered intensity of an X-ray beam hitting a sample as a function of incident and scattered angle, polarization, and wavelength or energy.

Materials that do not have long range order may also be studied by scattering methods that rely on elastic scattering of monochromatic X-rays:

Small angle X-ray scattering (SAXS) probes structure in the nanometer to micrometer range by measuring scattering intensity at scattering angles 2θ close to 0°

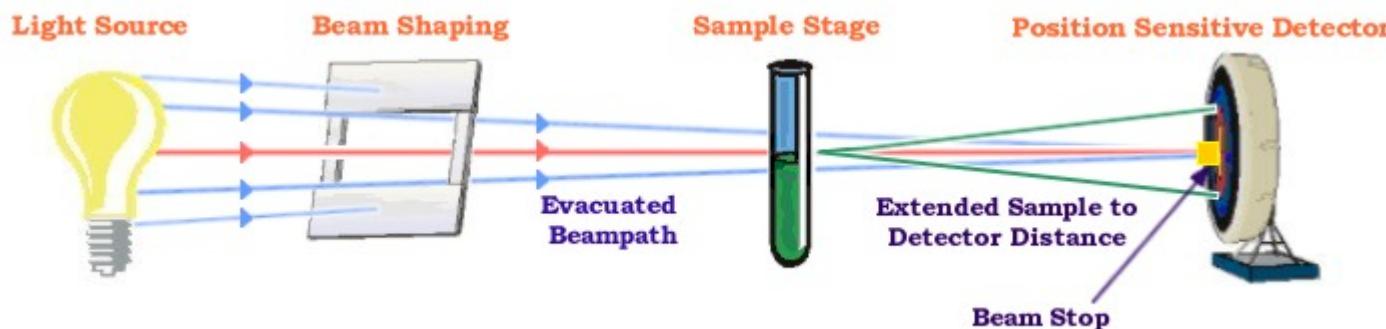
An Overview

Small-angle X-ray scattering (SAXS) is a small-angle scattering (SAS) technique where the elastic scattering of X-rays (wavelength 0.1 ... 0.2 nm) by a sample which has inhomogeneities in the nm-range, is recorded at very low angles (typically 0.1 - 10°). This angular range contains information about the shape and size of macromolecules, characteristic distances of partially ordered materials, pore sizes, and other data. SAXS is capable of delivering structural information of macromolecules between 5 and 25 nm, of repeat distances in partially ordered systems of up to 150 nm.

(Glatter, O.; O. Kratky (1982). Small Angle X-ray Scattering. Academic Press.
<http://physchem.kfunigraz.ac.at/sm/Software.htm>)

A SAXS Instrument

Conceptually, a SAXS experiment is simple: a sample is illuminated by X-rays and the scattered radiation is registered by a detector.



Until the 1970s, SAXS experiments were done on instruments equipped by laboratory X-ray tubes. Nowadays the X-ray source can be also a synchrotron light which provides a higher X-ray flux. Also neutrons can be employed.



A SAXS Instrument

The major problem that must be overcome in SAXS instrumentation is the separation of the weak scattered intensity from the strong main beam.

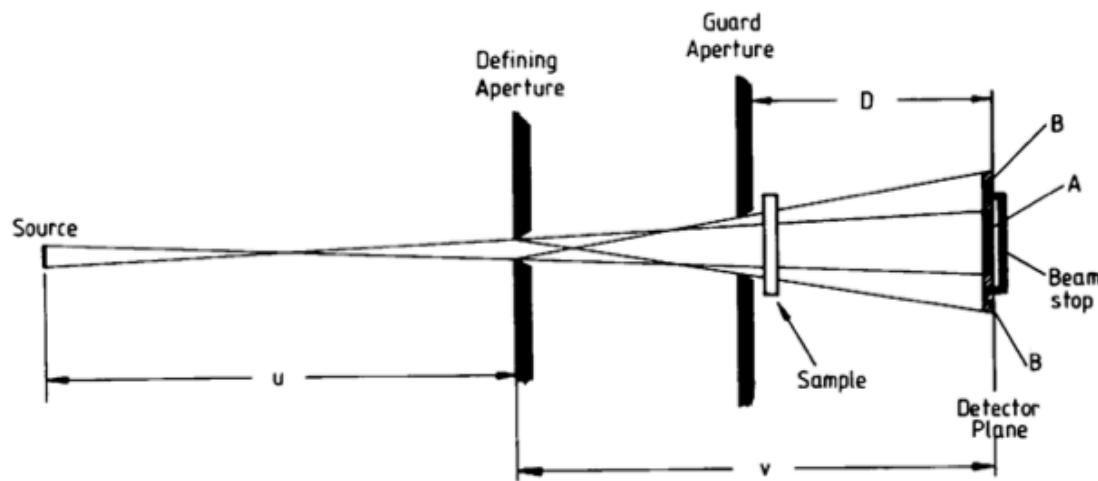


FIG. 1. Diagram showing the geometry of a point collimation camera. The angular resolving power is given by the diameter (A) of the collimated beam at the detector plane. The smallest angle of observation is determined by the diameter (B) of the penumbra produced by the guard aperture immediately in front of the specimen. The parameters u , v and D will be chosen to optimize the intensity for a given case (see, for example, Hendricks, 1978).

A SAXS Instrumentation

Laboratory SAXS instruments can be divided into two main groups:

1. Point-collimation instruments have pinholes that shape the X-ray beam to a small circular or elliptical spot that illuminates the sample. The scattered intensity is small and therefore the measurement time is in the order of hours or days in case of very weak scatterers.
2. Line-collimation instruments confine the beam only in one dimension so that the beam profile is a long but narrow line. The illuminated sample volume is much larger compared to point-collimation and the scattered intensity at the same flux density is proportionally larger. Thus measuring times with line-collimation SAXS instruments are much shorter compared to point-collimation and are in the range of minutes to hours.

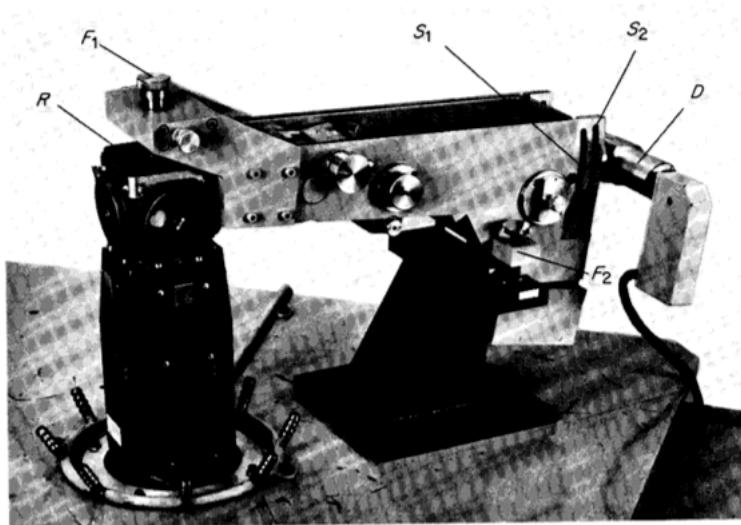


FIG. 6. Small angle camera.⁽¹¹⁾

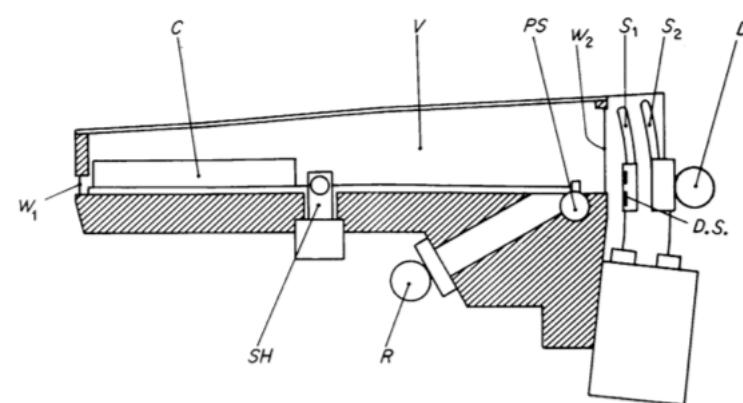
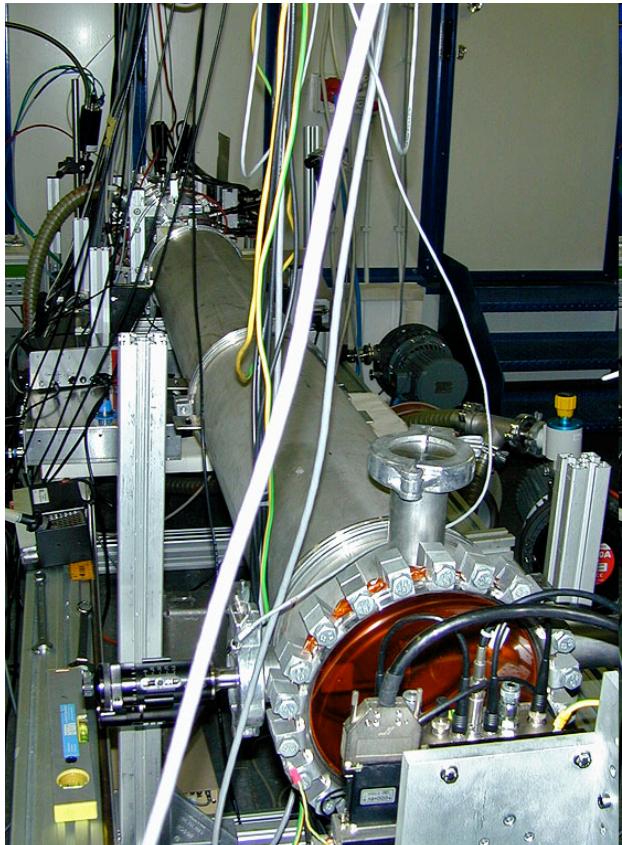


FIG. 7. Section through the small angle camera.⁽¹¹⁾

A SAXS Instrumentation

Synchrotron SAXS: Data can be collected in the minute/(milli)second time regime.



The camera length is variable between 1 and 10 m

SAXS Beamline

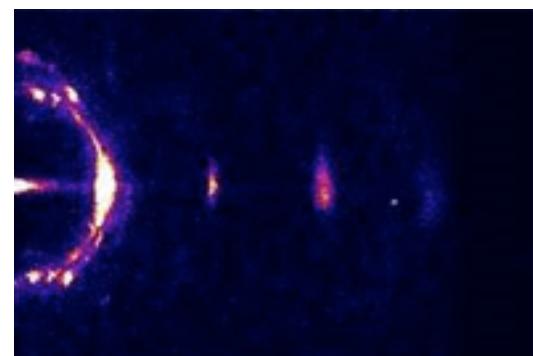
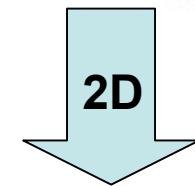
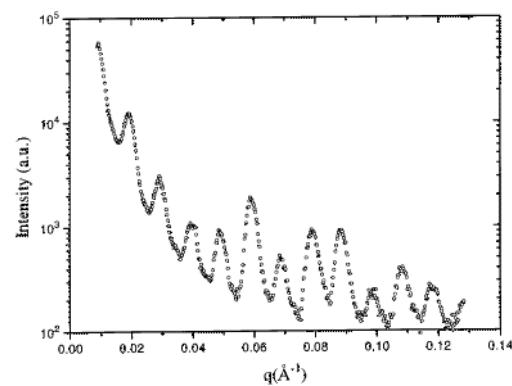
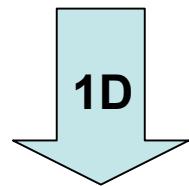
SAXIER: Small-Angle X-Ray Scattering Initiative for Europe

- The X33 beamline for biological small-angle scattering. EMBL/DESY, Hamburg, Germany
- The ID13 beamline ESRF, Grenoble, France
- The small- and wide-angle scattering beamline SWING. SOLEIL, France
- The Austrian small-angle scattering beamline BL 5.2 L. ELETTRA, Italy
- The non-crystalline diffraction beamline 2.1 in long geometry. SRS Daresbury, UK

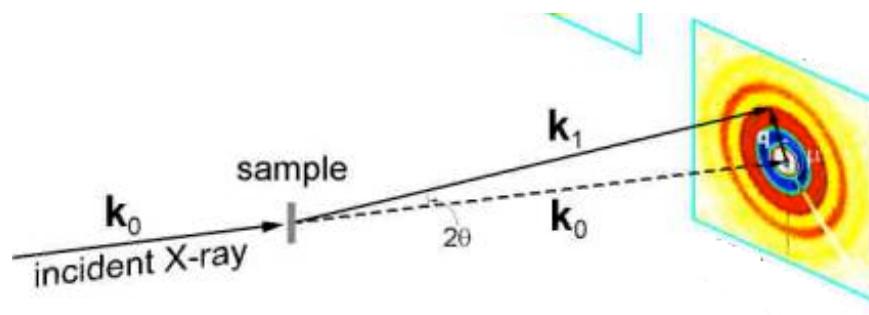


A SAXS Instrumentation

The detector and data acquisition system allows measurements in one- and two-dimensional manner.



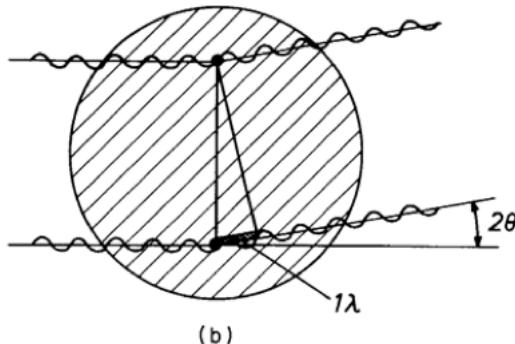
The scattering pattern contains the information on the structure of the sample.



??????

Why Small Angle?

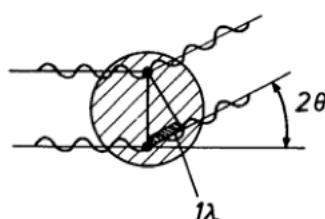
- 1) When an X-ray passes through an object it causes electrons to oscillate to the same frequency as that of the incident wave
- 2) These oscillating charges give rise coherent secondary electromagnetic waves with the same wavelength as that of the incident beam



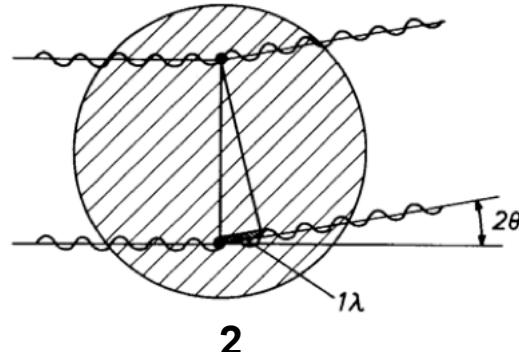
- 3) Any scattering process is characterized by a reciprocity law which gives an inverse relationship between object size and scattering angle

Why Small Angle?

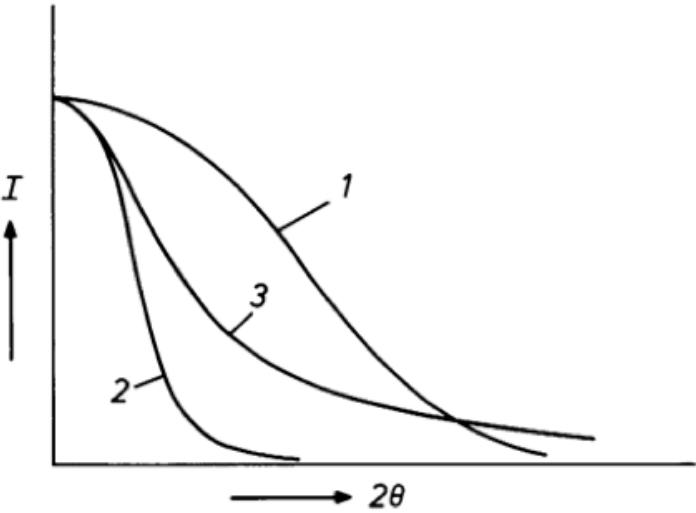
$$object \propto \frac{1}{angle}$$



1



2



Colloidal dimensions (between tens and several thousand Å) are enormously large compared to the X-ray wavelength (i.e. CuKa = 1.54Å)

Why Small Angle?

λ [nm]	Probed dimension	
	1°	0.1°
0.15	4.4nm	44nm
0.23	6.8nm	68nm
....
400	11μm	110μm

The SAXS technique is a *tool* which permits us to measure the scattered intensity at angle smaller than one degree

Diffraction < **SAXS** < Optical Microscope

SAXS vs. TEM

Transmission Electron Microscopy (TEM) cover the same dimensional range investigated by SAXS

The two techniques are complementary.

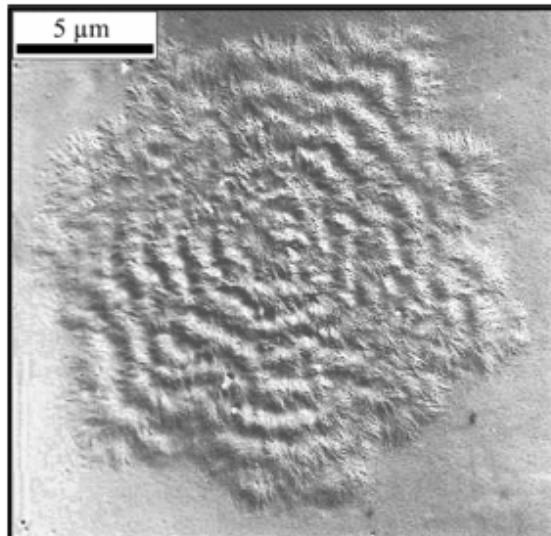
WHY?????

SAXS vs. TEM

Transmission Electron Microscopy (TEM) covers the same dimensional range investigated by SAXS

The two techniques are complementary

TEM good for:



Direct and detailed image

Local details

Local surface

Faithfully represents local complexities

TEM not good for:

Tiny image

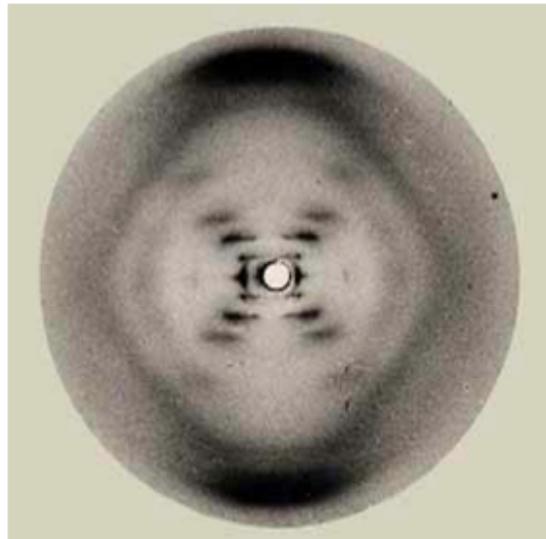
SAXS

Statistically significant average information

SAXS vs. TEM

Transmission Electron Microscopy (TEM) cover the same dimensional range investigated by SAXS

The two techniques are complementary



SAXS good for:

Global parameters and distribution

Different sample states(solid, wet or solutions samples...)

No destructive/artifacts in sample preparation

In situ transition study

SAXS vs. TEM

Transmission Electron Microscopy (TEM) cover the same dimensional range investigated by SAXS

The two techniques are complementary:

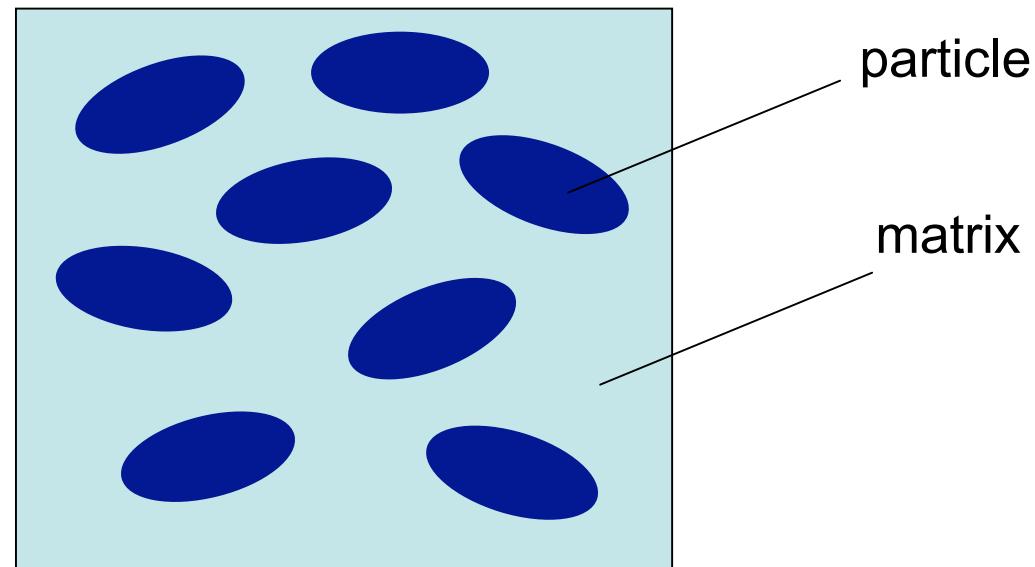
- TEM needs SAXS to obtain significant sampling and make quantitative statements
- SAXS needs TEM for clues about the shape of particles for use in data analysis

What is an object?

What SAXS ‘sees’ as objects are the spatial variations in the electron density inside the irradiated portion of matter

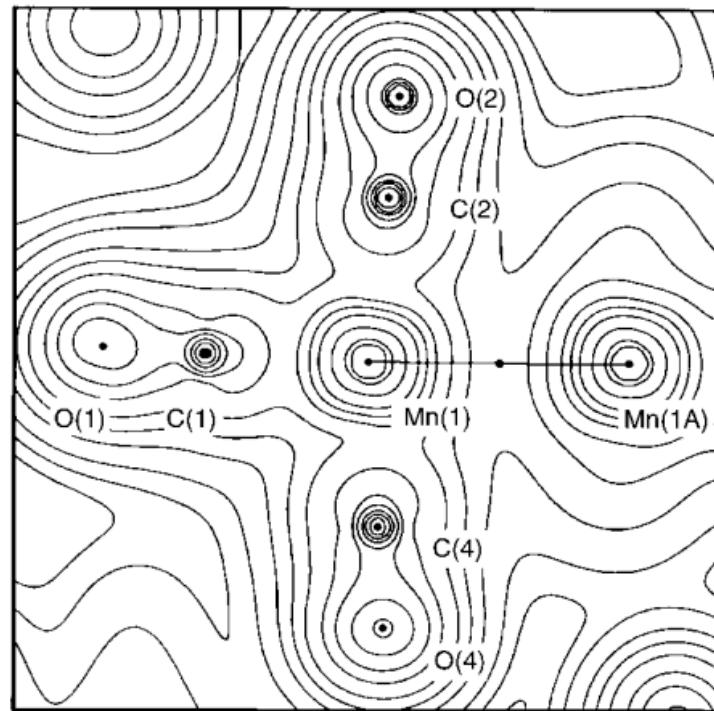
Electronic density difference is also called **contrast**

An example: A homogenous particle (i.e. a macromolecule) with a constant electron density dispersed in a matrix (i.e. a solvent) with a different electron density



What is an object?

On an atomic scale, the electron density continuously varies from higher values in the area surrounding an atomic position to lower values in the space between atoms

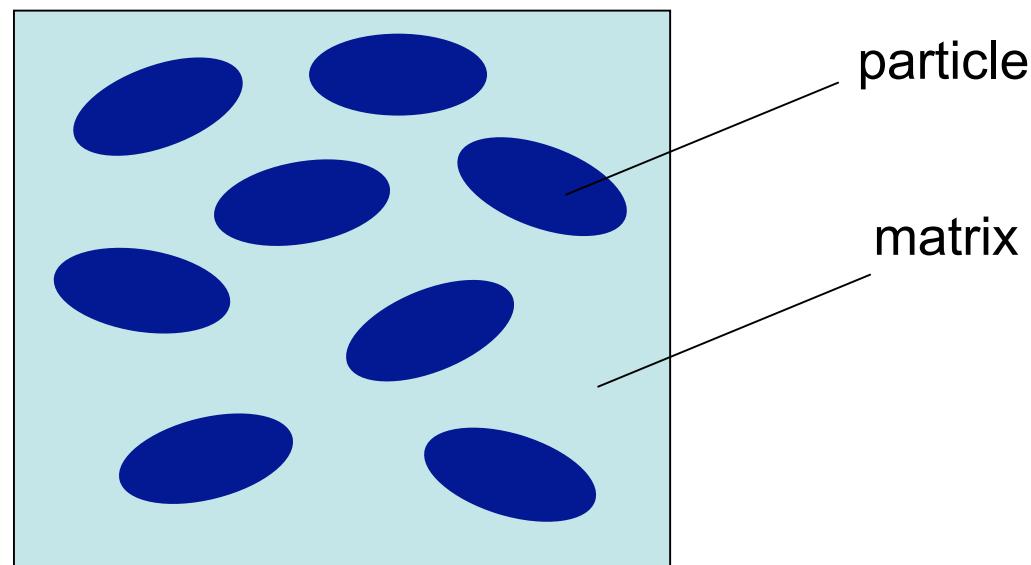


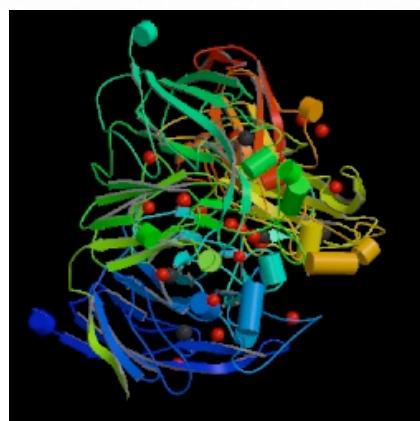
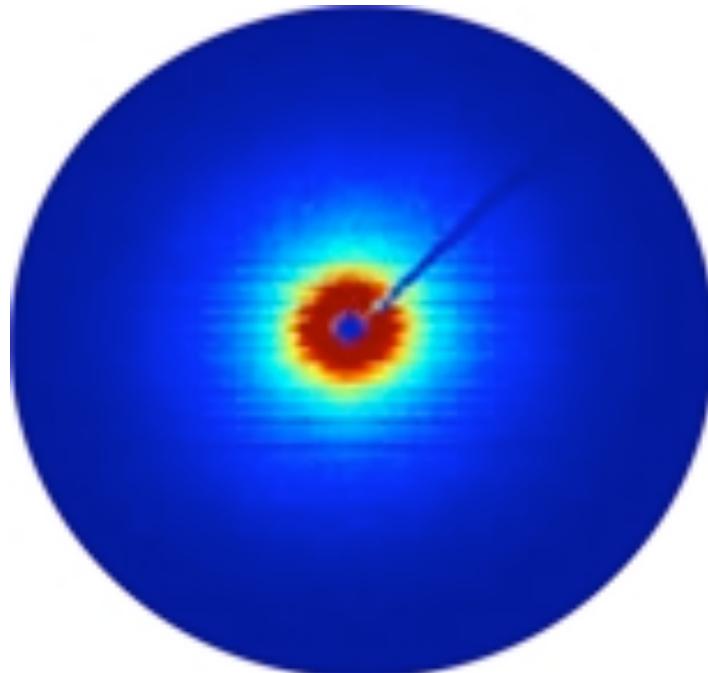
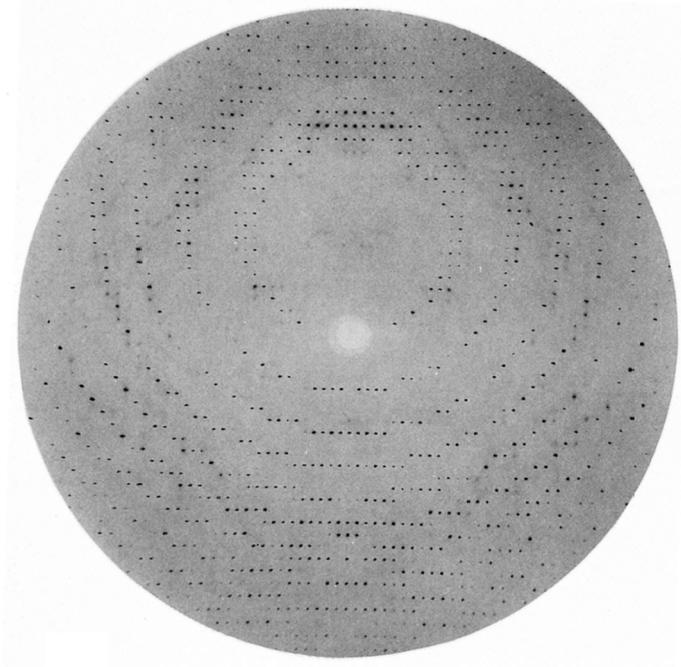
What is an object?

On an atomic scale, the electron density continuously varies from higher values in the area surrounding an atomic position to lower values in the space between atoms

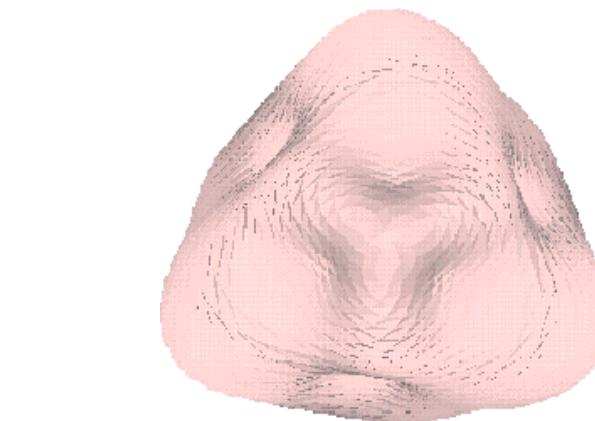
The SAXS explores dimensions of few nanometers. This implies that SAXS is not sensitive to electron density fluctuation on an atomic scale.

Consequently a portion of matter of homogenous composition is considered as a homogenous particle (a particle with constant electron density)





**Adman, E. T., Godden, J. W., Turley,
J Biol Chem 270 pp. 27458 (1995)**
Dr. Gianluca Croce



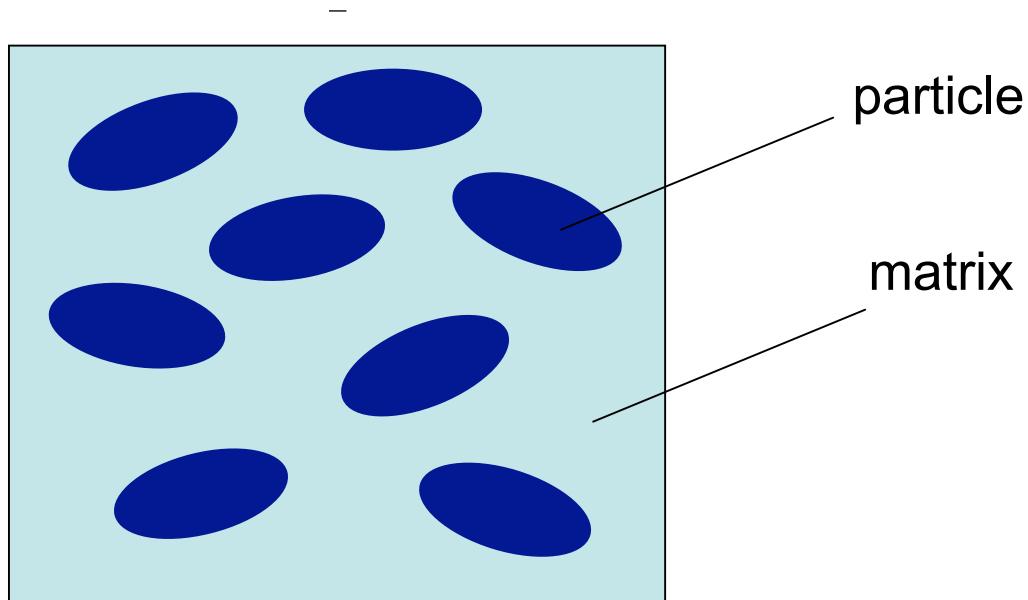
**Hao Q, et al., *Acta Crystallographica*
D55, (1), 243-246 (1999)**
21

What is an object?

The scattering is a good probe for matter with higher electron density contrast

The SAXS technique is good for:

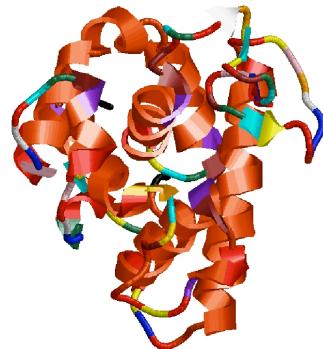
- amorphous particle
- crystalline particle
- homogeneous material with even voids(pores, blisters, cracks and crazes)



- SAXS is used for the determination of the microscale or nanoscale structure of particle systems in terms of such parameters as averaged particle sizes, shapes, distribution, and surface-to-volume ratio.
- 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.
- Not only particles, but also the structure of ordered systems like lamellae, and fractal-like materials can be studied.
- The method is accurate, non-destructive and usually requires only a minimum of sample preparation.
- Applications are very broad and include colloids of all types, metals, cement, oil, polymers, plastics, proteins, foods and pharmaceuticals and can be found in research as well as in quality control.

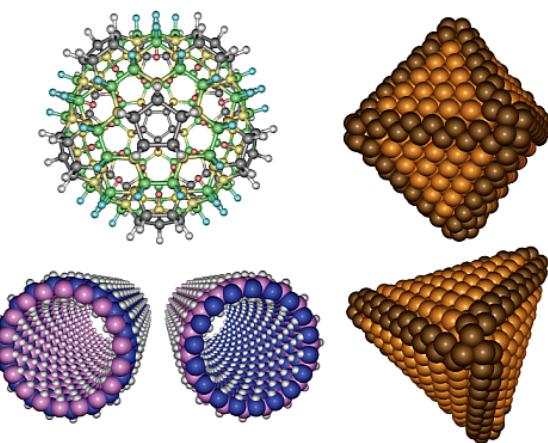
LIQUID

Protein; Pharmaceuticals.



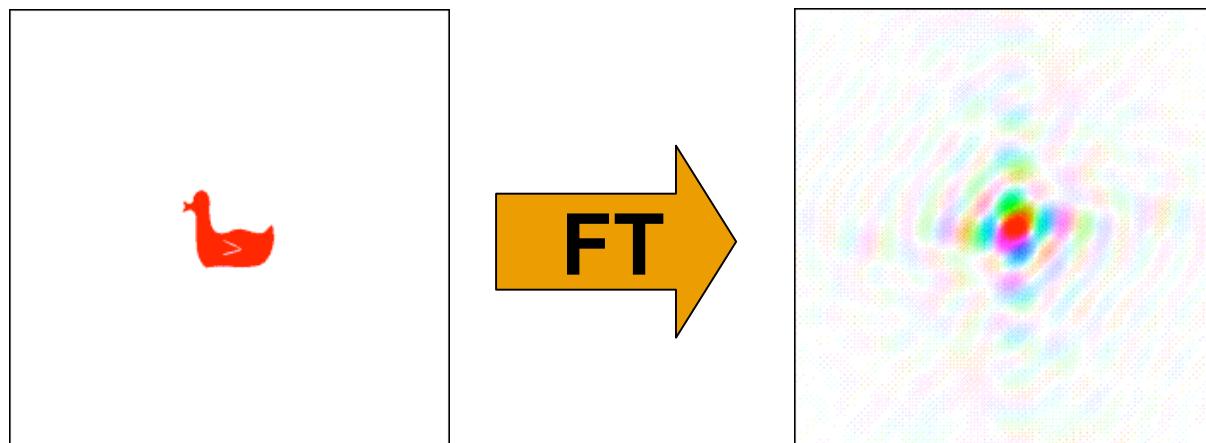
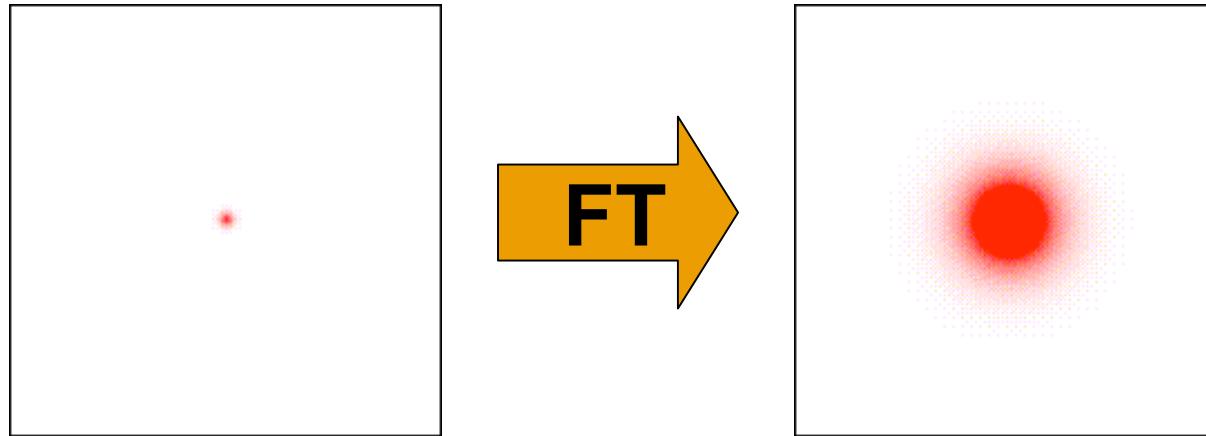
SOLID

Powder; Liquid Crystal;
Nano Materials; Polymer; Fiber.



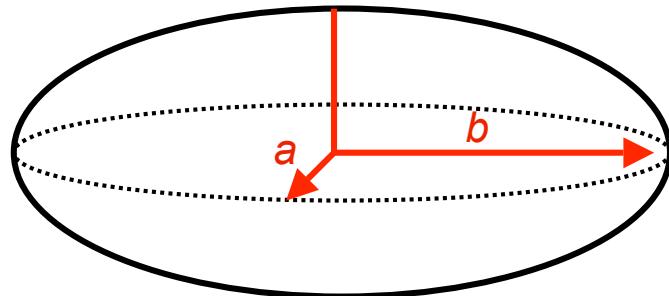
Single Particle Scattering

The scattering of a single particle in a matrix is *simply* its Fourier transform.



Single Particle Scattering

The Fourier transform can be calculated analytically for a large number of shape:



Ellipsoidal particle with constant orientation

The scattering intensity $I_0(h)$:

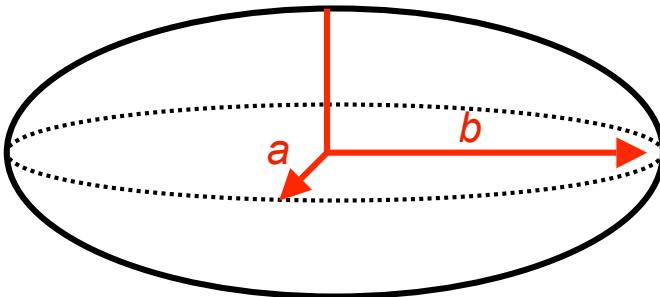
$$I_0(h, a, b) = \Delta\rho^2 \left(\frac{4}{3} \pi ab^2 \right)^2 \left(\frac{3\sin x - x\cos x}{x^3} \right)^2$$

Where:

$$x = \sqrt{a^2 h_1^2 + b^2 h_2^2}$$

$$\bar{h} \left(h = \frac{4\pi \sin \vartheta}{\lambda} \right) \quad \text{Scattering vector with } h_1 \text{ and } h_2 \text{ component on the detector plane}$$

Single Particle Scattering



$$I_0(h, a, b) = \Delta\rho^2 \left(\frac{4}{3} \pi ab^2 \right)^2 \left(\frac{3\sin x - x\cos x}{x^3} \right)^2$$

Electron Density Difference
(particle/matrix)

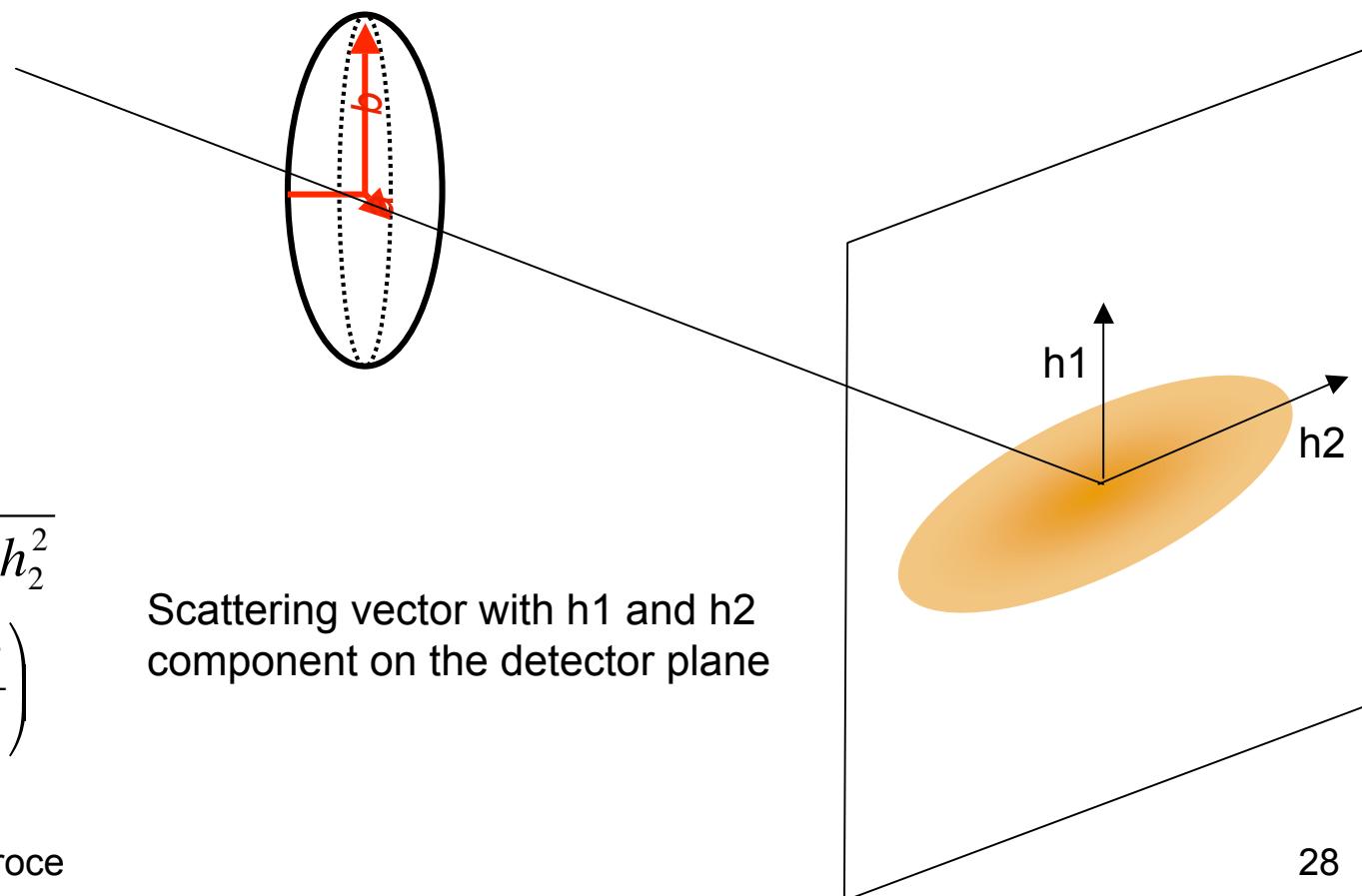
Volume of the ellipsoid

Bivariate function (surface) whose contour lines have an ellipsoidal shape oriented perpendicularly to the particle

Single Particle Scattering

$$\left(\frac{3 \sin x - x \cos x}{x^3} \right)^2$$

Bivariate function (surface) whose contour lines have an ellipsoidal shape oriented perpendicularly to the particle



$$x = \sqrt{a^2 h_1^2 + b^2 h_2^2}$$

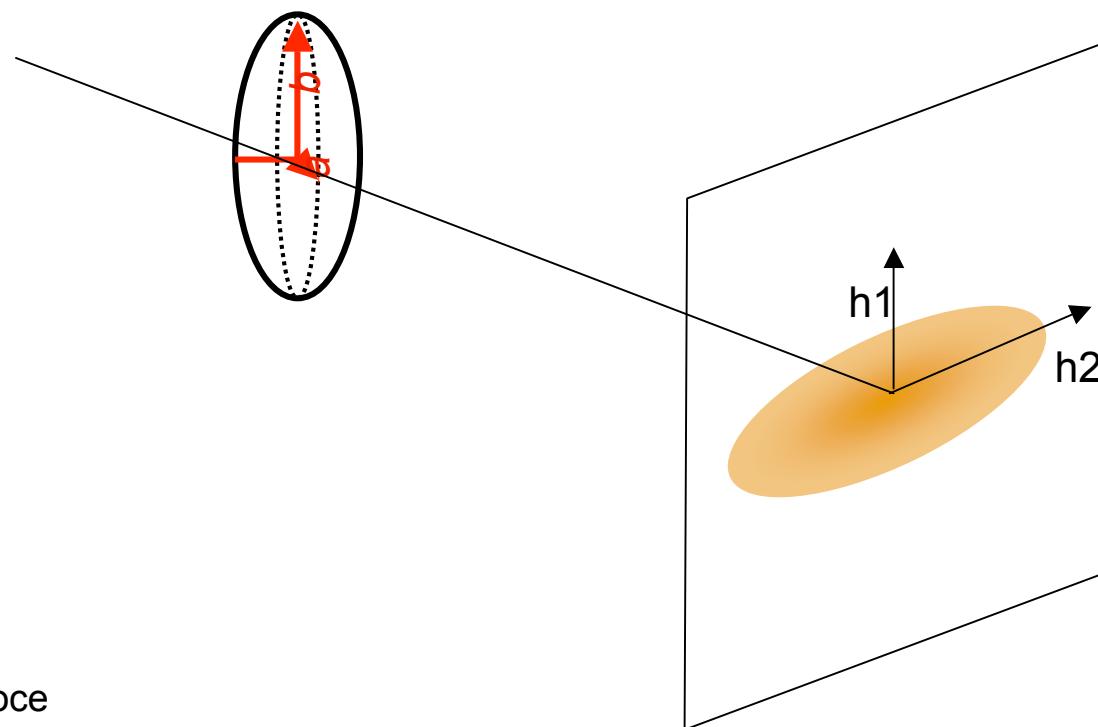
$$\bar{h} \left(h = \frac{4\pi \sin \vartheta}{\lambda} \right)$$

Single Particle Scattering

This last term is a consequence of the reciprocal law between the scattering particle and its scattering pattern

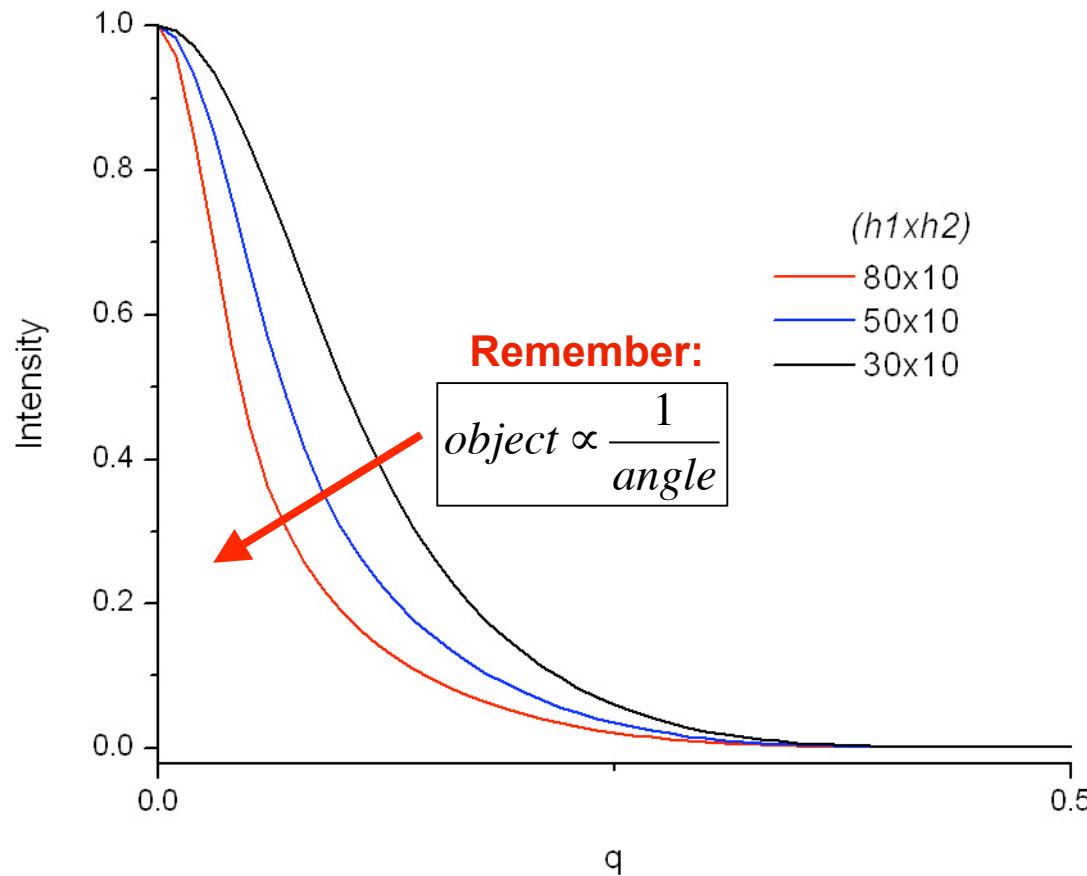
OR

Between a function and its Fourier transform



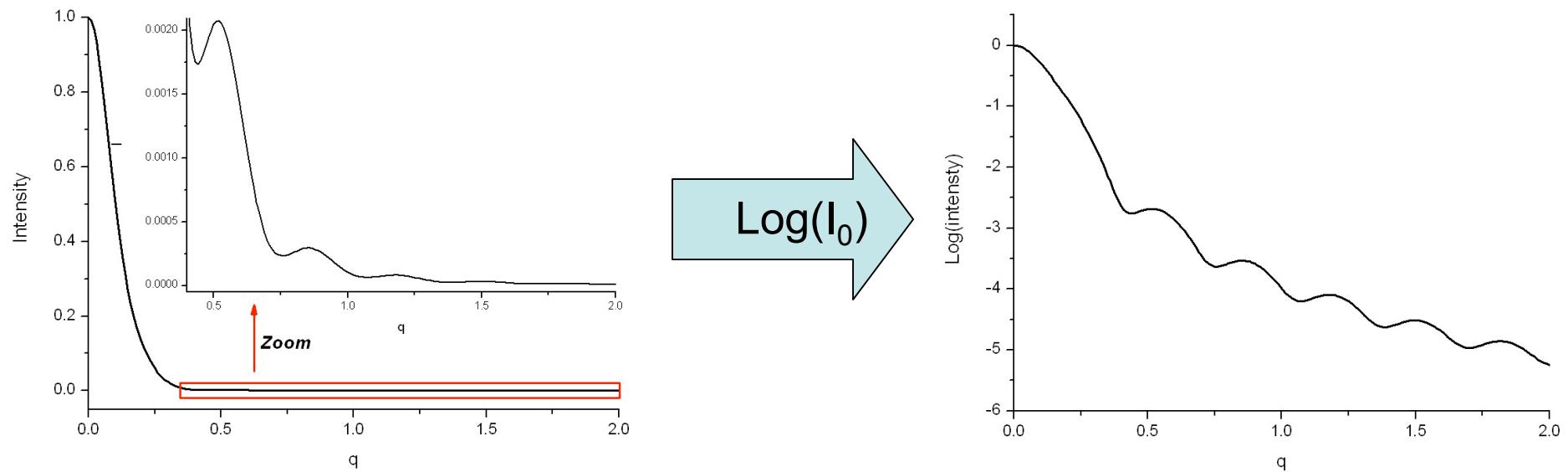
Single Particle Scattering

- Line sections of the scattering patterns (=linear detector) of an ellipsoid of different shapes
- The curves scattering decrease inversely to the vector $h1$



Single Particle Scattering

- A 1D scattering curve theoretically is composed by a central peak with a series of rapidly damped peaks of much smaller intensity
- For simplicity is useful to represent the data in a logarithmic scale to better appreciate the small intensity peaks



Single Particle Scattering

- Scattering equation of particles of different shape:

Sphere: radius R

$$I(q) = N n^2 \{3(\sin qR - qR \cos qR)/(q^3 R^3)\}^2$$

$$R_g = R/1.29 = R\sqrt[4]{(3/5)}$$

Rod: Length 2H; Diameter 2R

$$I(q) = N n^2 \pi \exp(-q^2 R^2 / 4) / (2qH)$$

$$R_{g_{overall}} = R^2/2 + H^2/3$$

Disk: diameter 2R; thickness 2H

$$I(q) = N 2n^2 \exp(-q^2 H^2 / 3) / (q^2 R^2)$$

$$R_{g_{overall}} = R^2/2 + H^2/3$$

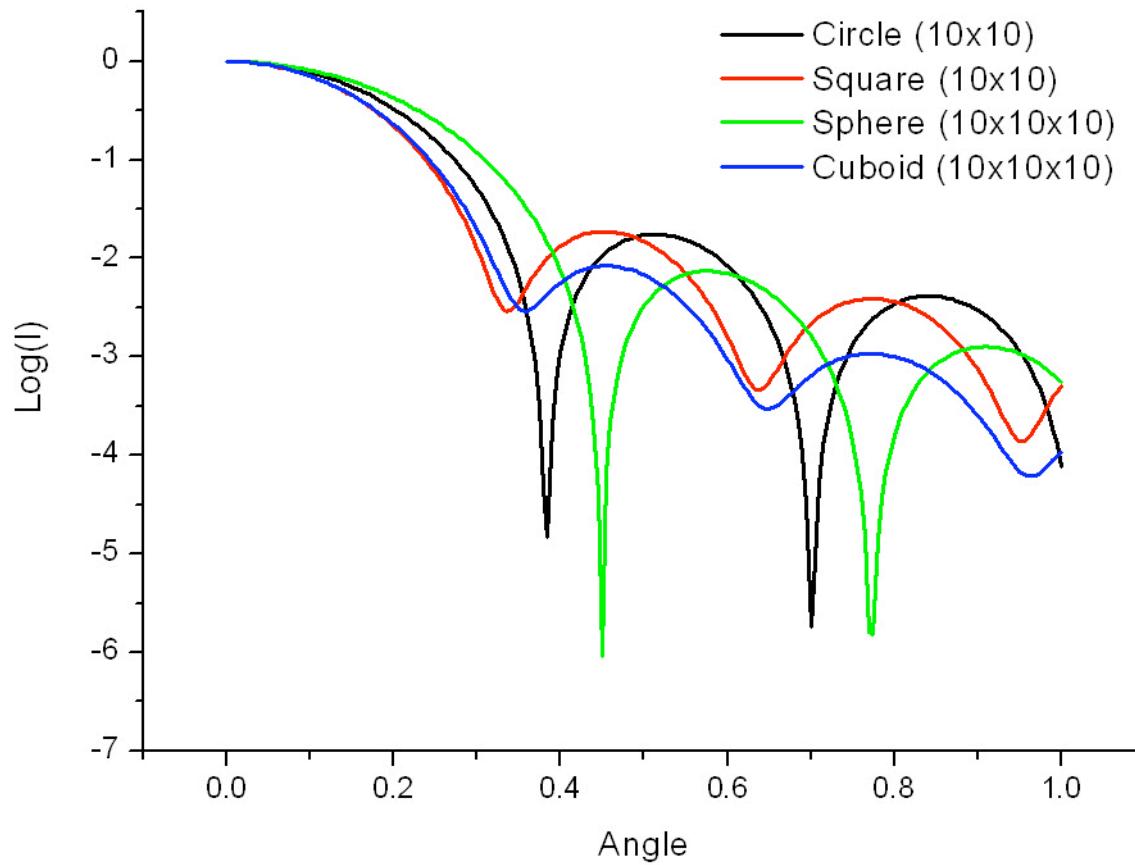
Gaussian Polymer Coil: n persistence units; l persistence length

$$I(q) = I_0 \{2(Q - 1 + \exp(-Q)\}/Q^2$$

$$Q = q^2 R_g^2$$

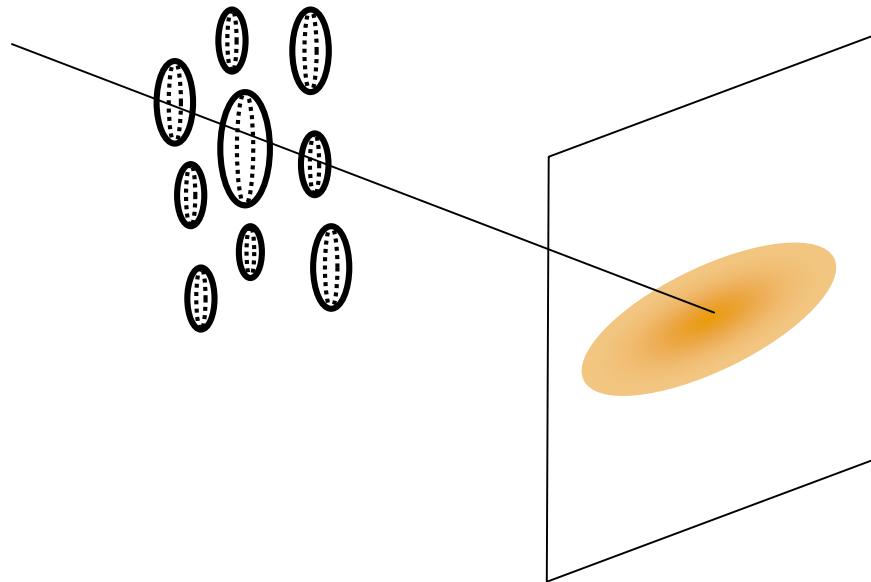
Single Particle Scattering

- 1D scattering curve of particles of different shape and equal dimensions



Group of Particle Scattering

In a real case we normally study system composed by many particles dispersed in a matrix (i.e. a solvent)



A *dilute system* of ellipsoidal particle with the same orientation:

- **Same dimension:** a simple sum of the scattering of each single particle
- **Different dimension:** multiply the function for the scattering of a single particle by the distribution of sizes

Group of Particle Scattering

What does it mean “Multiply the function for the scattering of a single particle by the distribution of sizes”?

$$I_0(h,a,b) = \iint D(a,b) I_0(h,a,b) da db$$

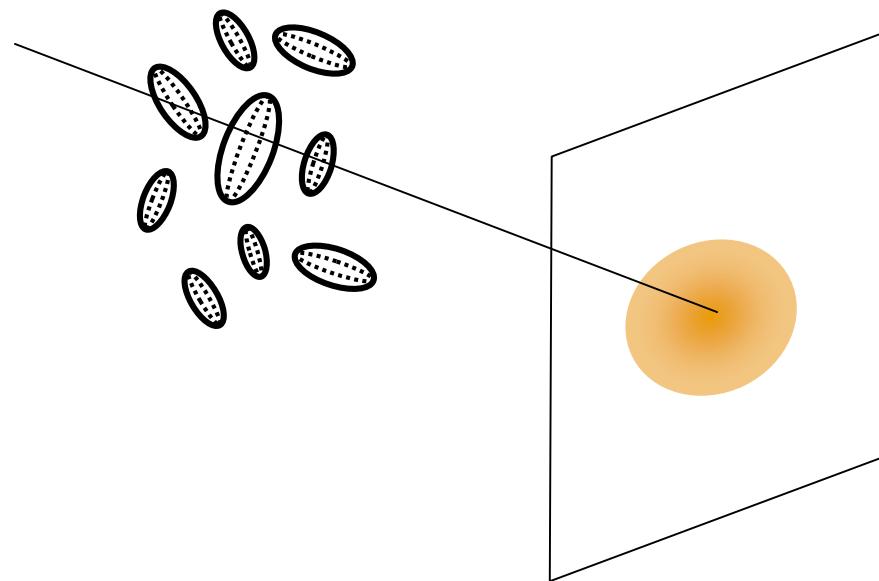
Function of the semi-axes of the ellipsoid; a and b parameters control the dimensions of the particles

Equation with described the scattering for a single ellipsoid:

$$I_0(h,a,b) = \Delta\rho^2 \left(\frac{4}{3} \pi a b^2 \right)^2 \left(\frac{3 \sin x - x \cos x}{x^3} \right)^2$$

Group of Particle Scattering

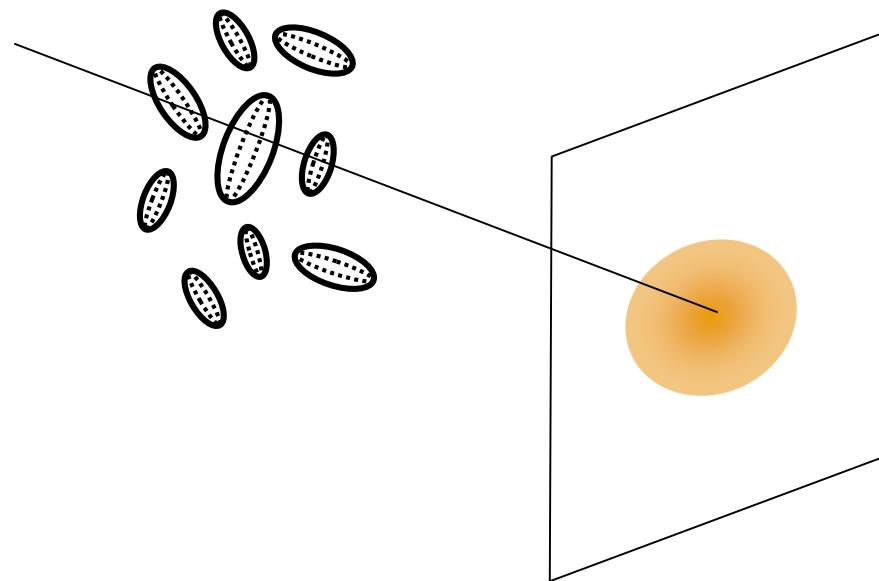
If the orientation of the particles vary or if their orientation changes over time (random orientation):



The scattering is averaged in all directions and the pattern will be isotropic

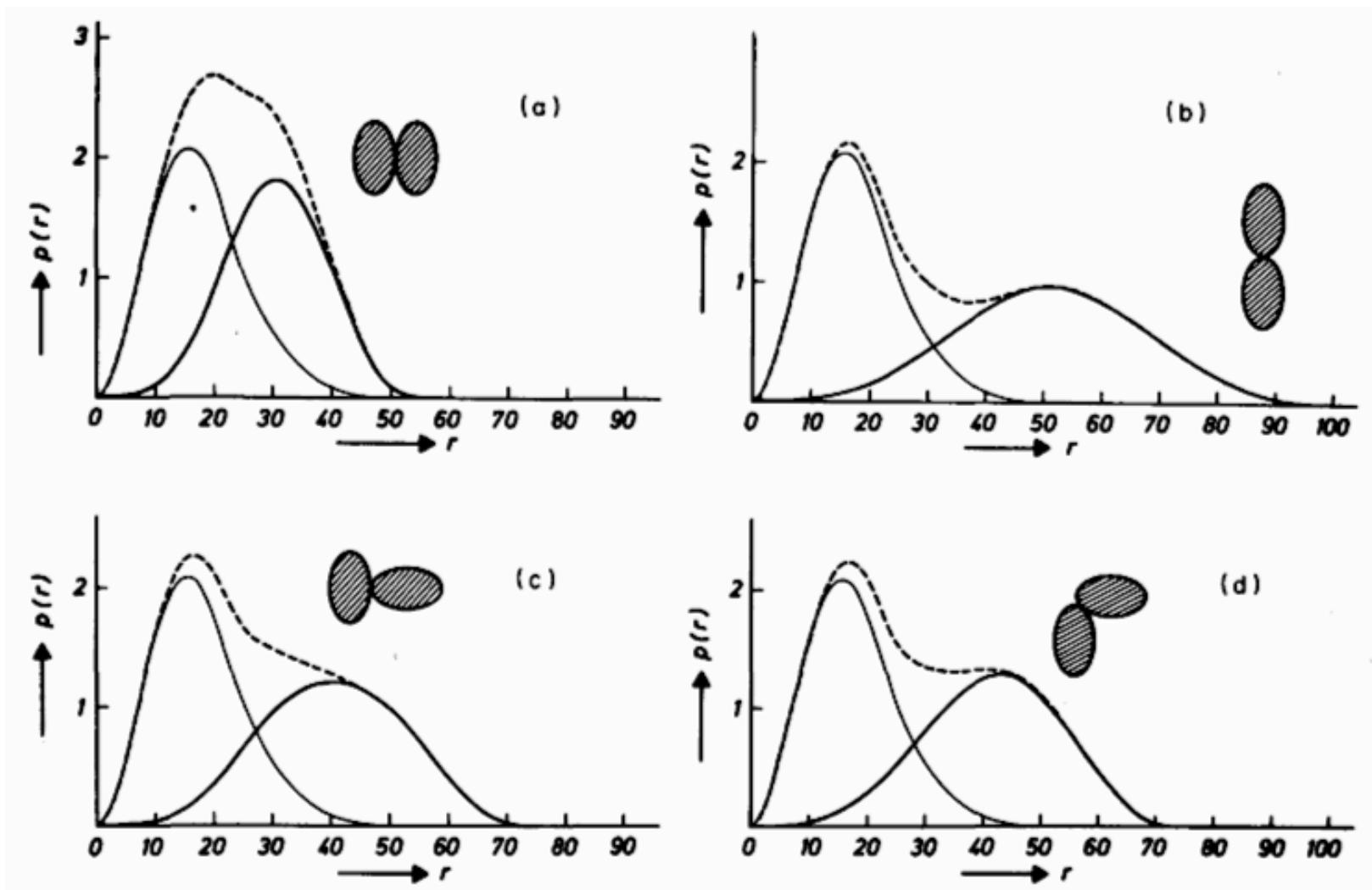
Group of Particle Scattering

In this ‘real’ situation, a distribution of directions should be introduced for mimic the scattering pattern $I(h,a,b)$



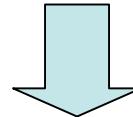
Obviously the information on particle shapes and dimensions is mixed and it's necessary to make assumptions about their shape or size distribution

Group of Particle Scattering

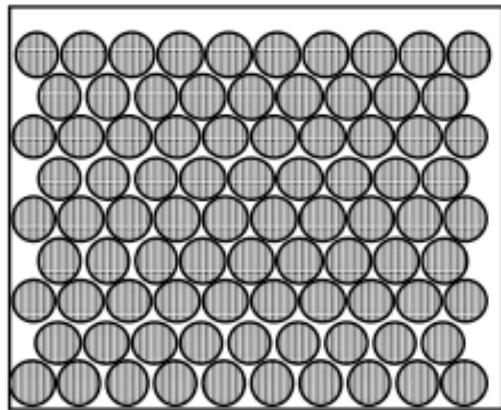


Group of Particle Scattering(Densely Packed)

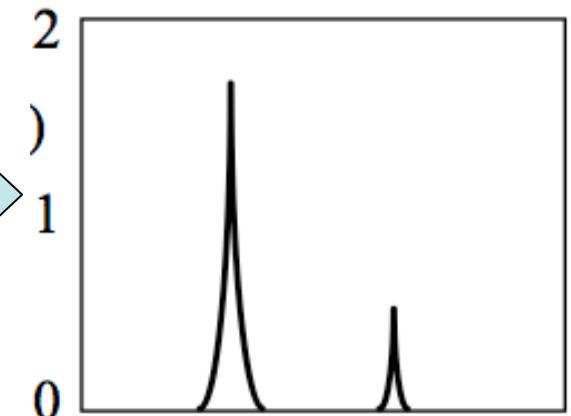
When particles are very close one to another



They cause a constructive interference similar to the Bragg peaks (diffraction).



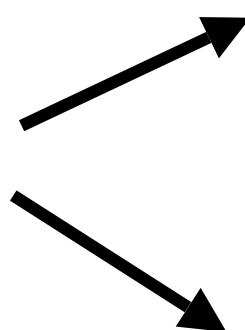
If these distances have some degree of regularity



Data Analysis

- A SAXS signal contain information about shape and size particles (also distances)
- These information are confused in a SAXS pattern
- These contributions are impossible to separate without external information (i.e. particle shape, a certain distribution, etc...)

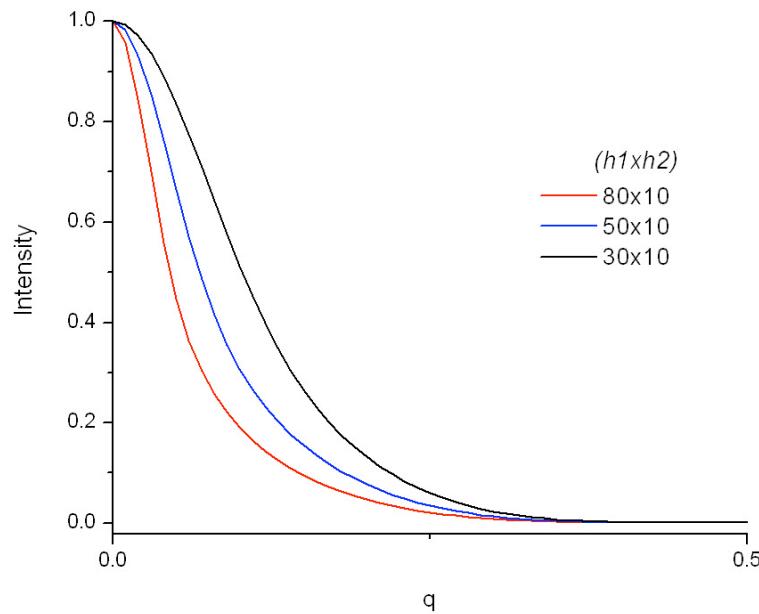
Data Analysis Manipulation



- Retrieve particles information processing experimental data by means of mathematical transformations to a assumed shape
- From an assumed particle distribution, calculate the scattering intensity in order to try to fit the experimental data

Guinier Approximation

At smallest angle, the scattering curve of a dilute system (similar size particles) can be approximated:



$$I(h) \propto e^{-aR_g^2 h^2}$$

Gaussian Function

Where:

Rg is called Gyration Radius

a is a factor depending on particle orientation(i.e. random orientation a=1/3)

Guinier Approximation

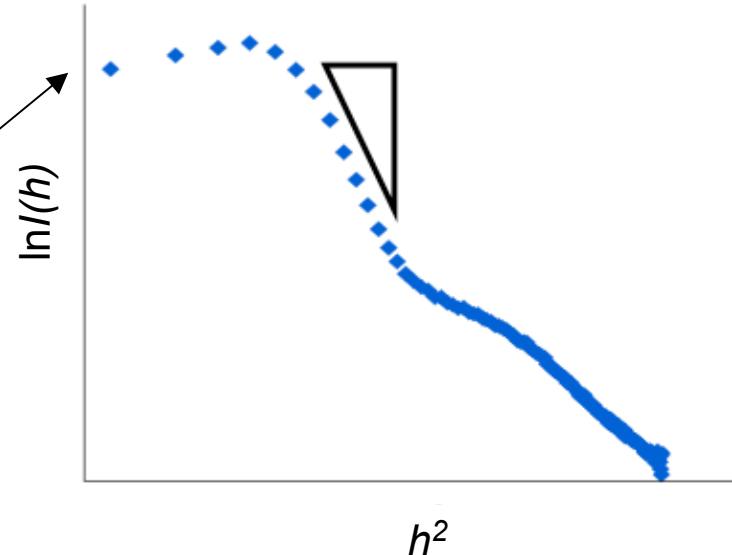
The Gyration Radius is defined in analogy to the radius of inertia in mechanics.

R is defined as the mean square distance from the centre of gravity ($R=\sqrt{\langle r^2 \rangle}$) where the role of the 'mass' is played by the electrons.

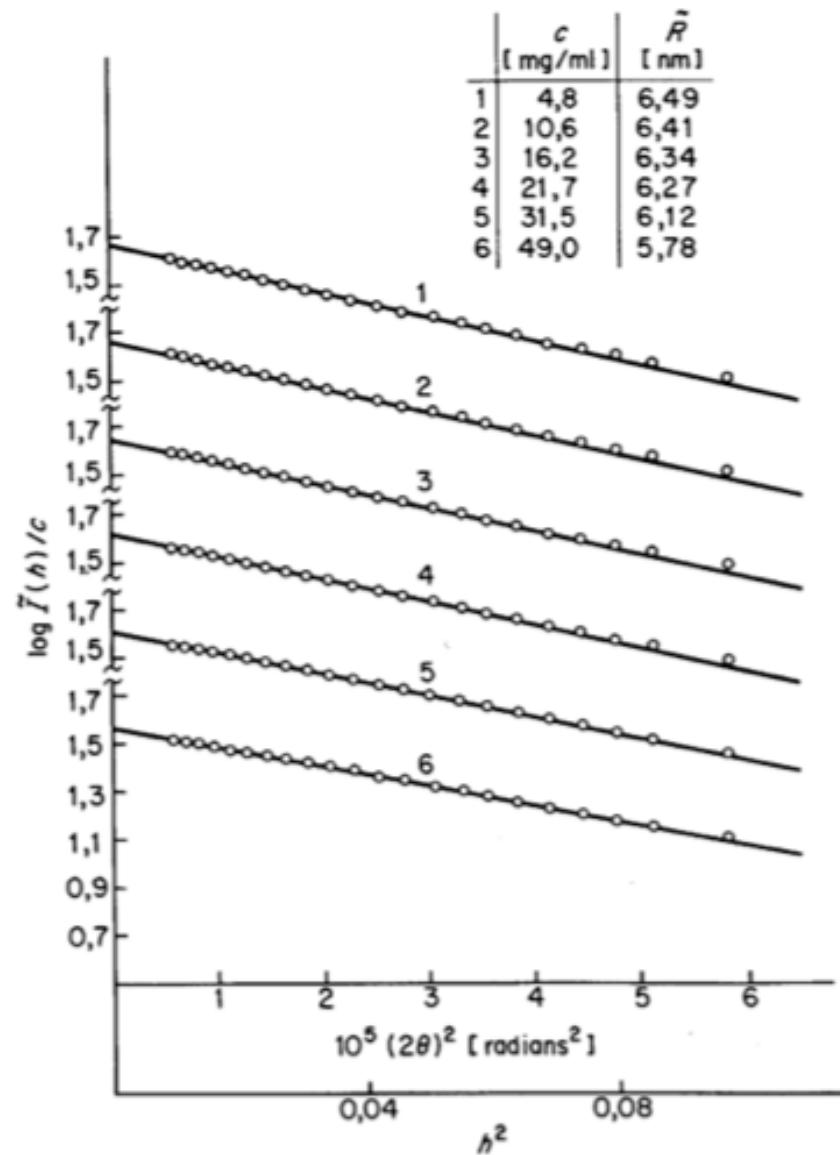
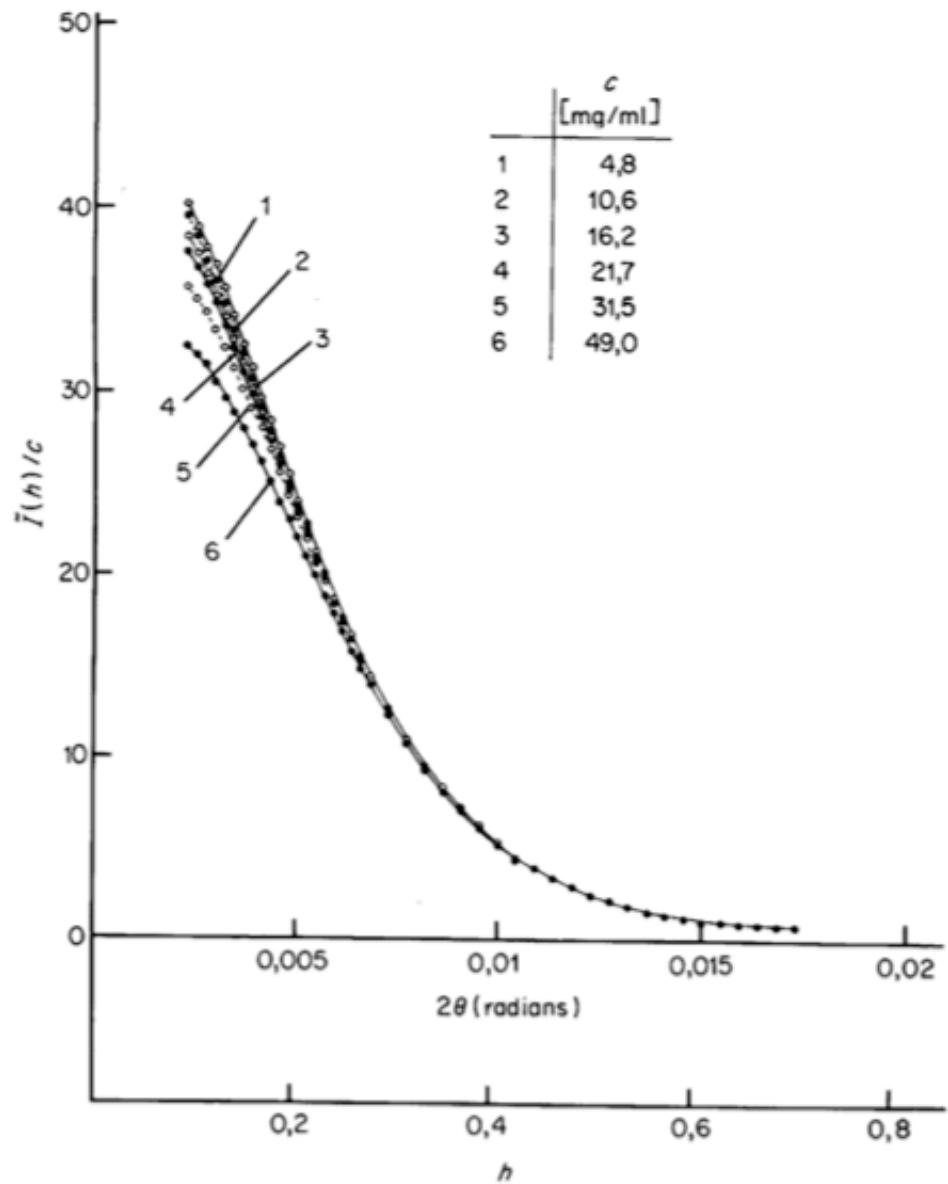
The equation of randomly oriented particle ($a=1/3$) is:

$$I(h) = I(0)e^{-\frac{-R^2 h^2}{3}}$$

R_g can be calculated from the slope of the strain region of the plot

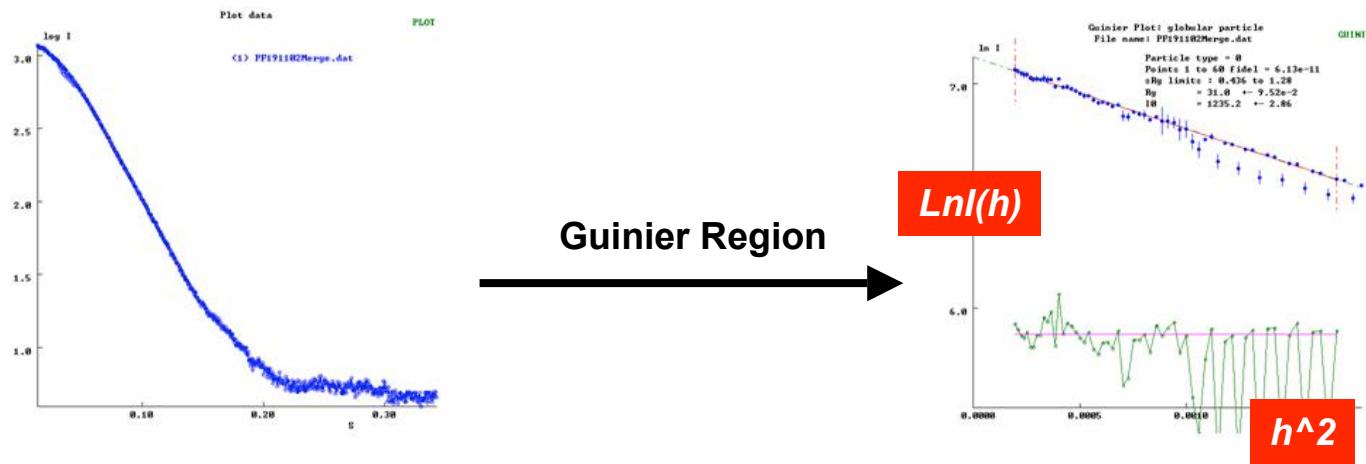


Guinier Approximation



Guinier Approximation

- The R_g is a parameter doesn't provide any geometrical information of particles.
- It's always necessary to consider an '*a priori*' geometry (shape) of the particles.
- If the particle shape is defined, the Guinier approximation provides a good indication of average size particles.

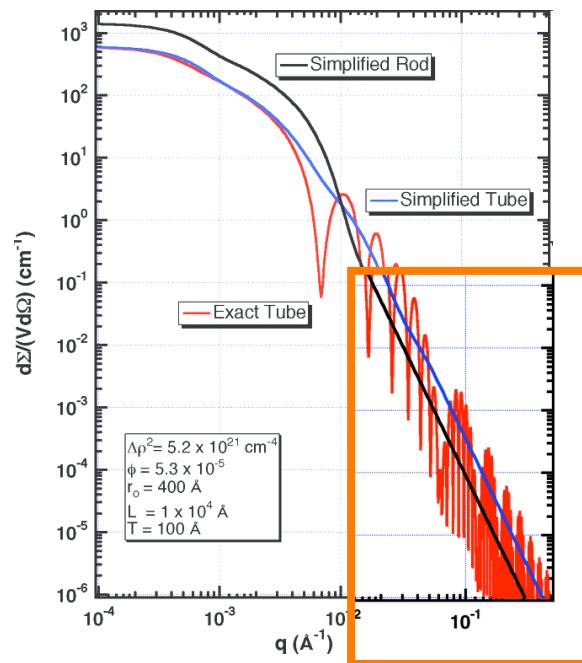


The Guinier approximation works well when, in a dilute system, the particles preserve the same shape with also different dimension (in a limited range)

If the particle dimensions are drastically different may occur to experimentally observe two definite Guinier region (determinable separately)

Porod Law

- Examining the queues of a SAXS signal, Porod observed that in most case the data follow an asymptotic behavior with a fourth powerlaw decay
- This behavior is expected for so-called two-phase system (when the el. density ρ can assume only two value: ρ_1 and zero)



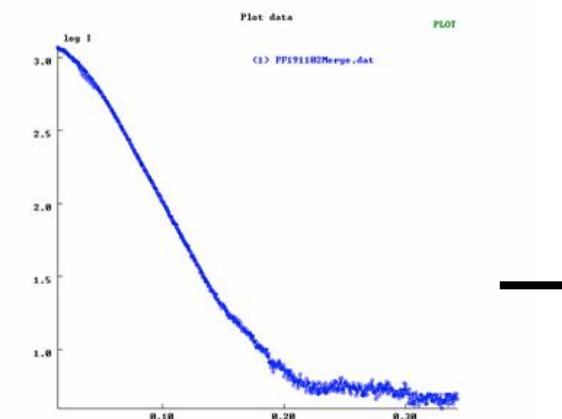
Porod Law

Mathematically for large value of h :

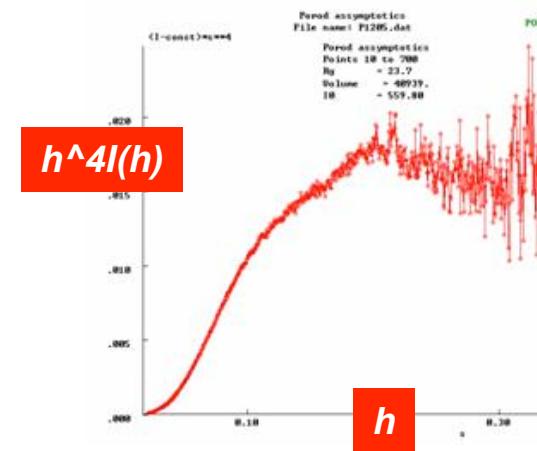
$$I(h) = \frac{2\pi}{h^4} S (\rho_1 - \rho_0)^2$$

Where S represents the surface area of the particles and may be determined from experimental data.

In an ideal case (spherical particle with $\rho=\rho_1; 0$) the Porod Plot trends to a plateau. This constant value is proportional to the surface area at the interface.



Porod Plot



Integral Quantities

Porod also demonstrated that normalizing the intensities (Absolute Intensity), the following relation for a two-phase system is possible:

$$\frac{1}{V} \int_0^{\infty} h^2 I(h) dh = v(1-v)(\rho_2 - \rho_1)^2$$

V = scattering volume

$\rho_1; \rho_2$ = electronic density of two phases

$v; 1-v$ = volumetric fraction of two phase

Scattering Power

This quantity is calculable from the experimental data if it's possible to extrapolate them to zero (Guinier) and to infinity (Porod)

This equation permit to estimate the volumetric fraction knowing the electronic density, and viceversa, of a two-phase system.

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