Solution X-ray Scattering from Biological Macromolecules

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Oléron- 4 Juin2018

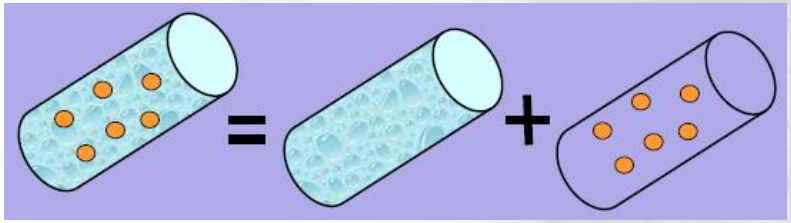
INTRODUCTION

Principles of Small Angle X-ray Scattering in solution

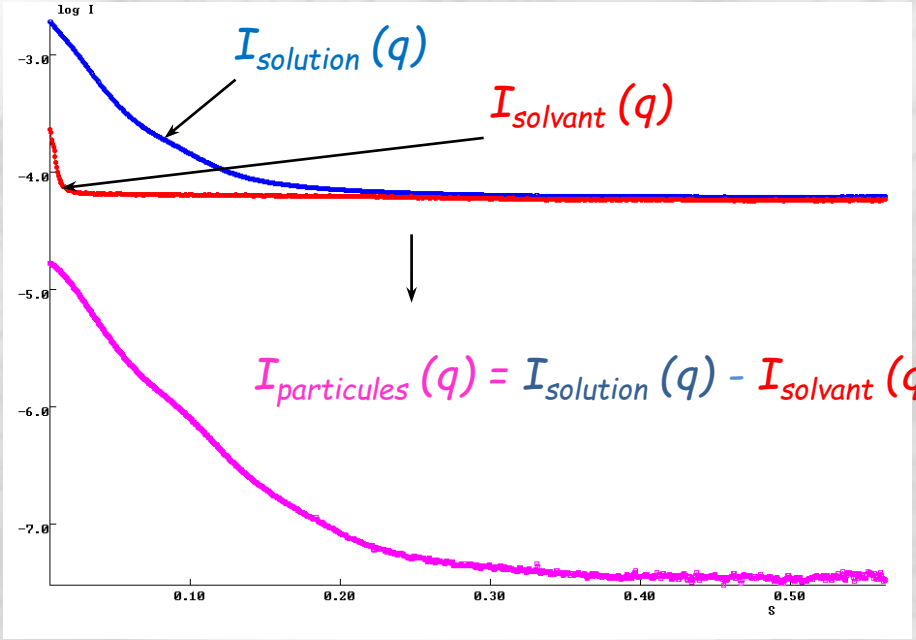
- **Limits**
  - spherically averaged information → low resolution
  - non unicity of the solution
  - does not distinguish elements in a mixture
- **Advantages**
  - solution (no crystal) → kinetics, titration, T°, P
  - relatively easy to carry experiments
  - can be checked against atomic models

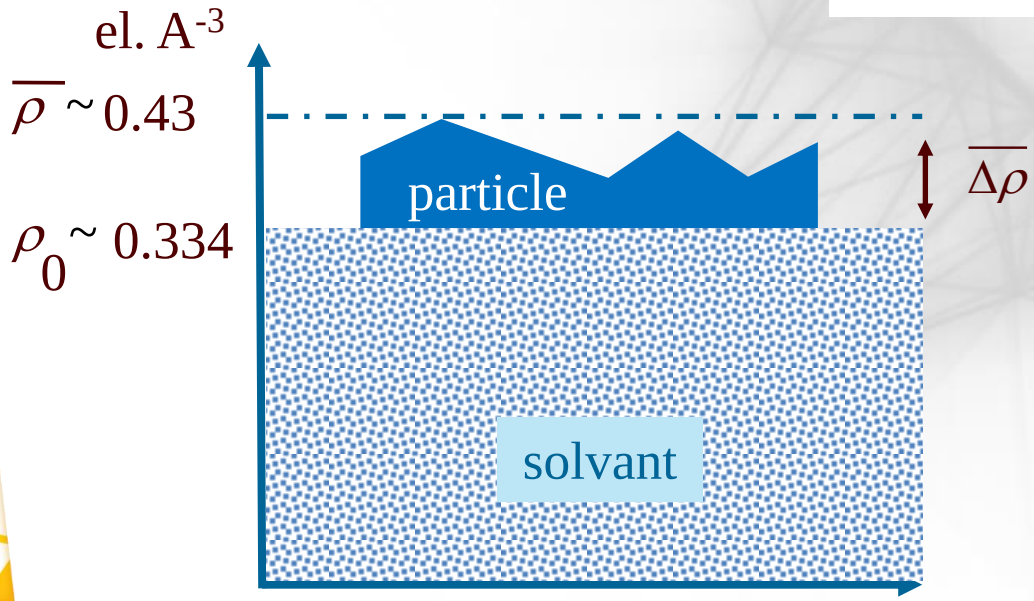
Principles of Small Angle X-ray Scattering in solution

Method sensible to the difference between the electronic density of the particule and the solvent (contrast)



$I_{solution}(q)$ $I_{solvant}(q)$ $I_{particules}(q)$





In a matrix, what contributes to scattering is the ***contrast*** of electron density between the particle and the matrix $\Delta\rho(\mathbf{r}) = \rho_p(\mathbf{r}) - \rho_0$ that may be **very small** for biological samples.

Principles of Small Angle X-ray Scattering in solution

- **Monodispersity**
  - Yes ← Identical particles
  - No ← Size and Shape polydispersity

$$i_i(q) = i_1(q)$$

- **Ideality**
  - Yes ← No correlations between particles positions (No short-range or long-range interactions)
  - No ← Correlations between particles positions (Existence of short-range or long-range interactions)

$$I(q) = \sum_{i=1,N} i_i(q)$$

$$I(q) = N i_1(q)$$

Principles of Small Angle X-ray Scattering in solution

Structural information obtained from scattering curve

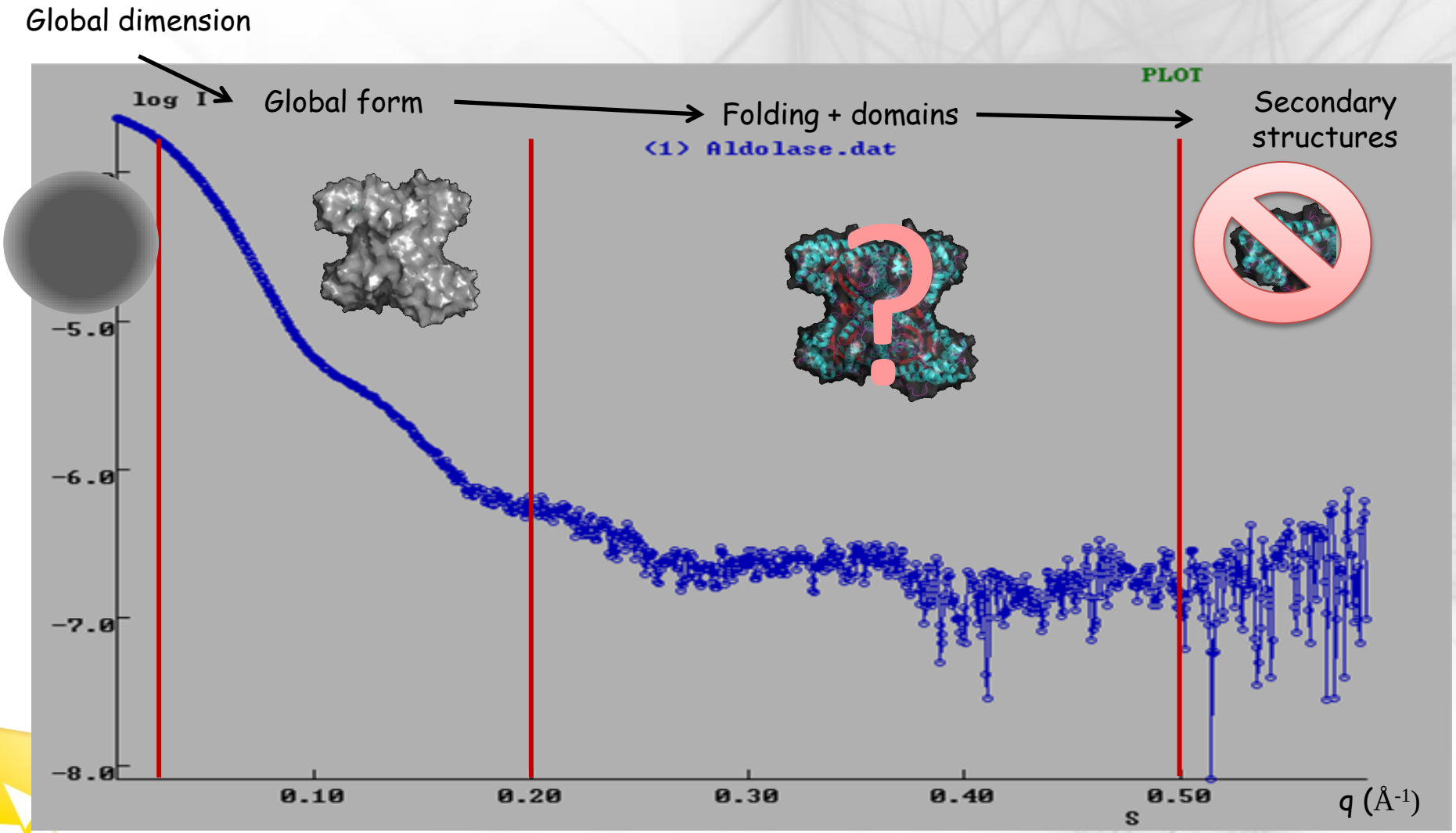
- ***Biophysical informations calculated directly from the SAXS curve***
  - biophysical parameters : size, shape, fold of the object (Rg, P(r), Kratky, …)
  - molecular weight, oligomerization state and volume (I_0, Porod, …)

- ***3D structural informations***
  - low resolution molecular shape calculation with ab initio method
  - comparison with high resolution model
  - molecular modeling of unstructured missing part
  - molecular modeling rigid body of complex

SAXS data compatible model
NOT
a structure

DATA ANALYSIS

What may solution scattering yield?



Data Analysis

- Guinier Analysis
- Kratky plot : why is it so interesting ?
- « Real-space SAXS » : Distance correlation function P(r)

Data Analysis: Guinier law

Close to q=0, the scattering intensity of a particle can be described by a Gaussian curve.
The validity domain actually depends on the shape of the particle and is around $\mathbf{q_{max} < 1.3 / Rg}$ for a globular shape.

Prof. André Guinier
1911-2000
Orsay, France

$$I(q) = I(0)\exp\left(\frac{-q^2 Rg^2}{3}\right)$$

Extrapolated intensity at origin

Radius of gyration

Guinier law, in Log scale :

$$\ln[I(q)] = \ln[I(0)] - \frac{q^2 Rg^2}{3}$$

The Guinier law is equivalent of a linear variation of Ln(I(q)) vs q^2 (Guinier plot).
Linear regression on the experimental Guinier plot directly provides R_g and I(0).

Data Analysis: Guinier law: Mass estimation

$$I(Q) = I(0)\exp\left(\frac{-Q^2 Rg^2}{3}\right)$$

Absolute Unit : cm^{-1}

Classical electron radius

$$I(0) = \frac{c \cdot M \cdot r_0^2}{N_A} \cdot \left[v_p\left(\rho_{prot} - \rho_{buf}\right)\right]^2$$

$$Rg^2 = \frac{\int_V r^2 \Delta\rho_{prot}\left(\vec{r}\right)d\vec{r}}{\int_V \Delta\rho_{prot}\left(\vec{r}\right)d\vec{r}}$$

Mass concentration

Electronic density contrast

Protein specific volume

I(0) gives an independent estimation of the molar mass of the protein **(only if the mass concentration, c, is precisely known …)**

Rg depends on the volume AND on the shape of the particle

For globular proteins : $R_g\,(Å) \approx 6.5 * M^{\frac{1}{3}}, M\ in\ kDa$

For unfolded proteins : $R_g\,(Å) \approx 8.05 * M^{0.522}$

Bernado et al. (2009), Biophys. J., 97 (10), 2839-2845.

Typically :
M (kDa) = 1500 * I0 (cm^{-1}) / C (mg/ml)

Data Analysis: Guinier law

Guinier analysis

R_g → size

I(0) → mol mass / oligomerisation state

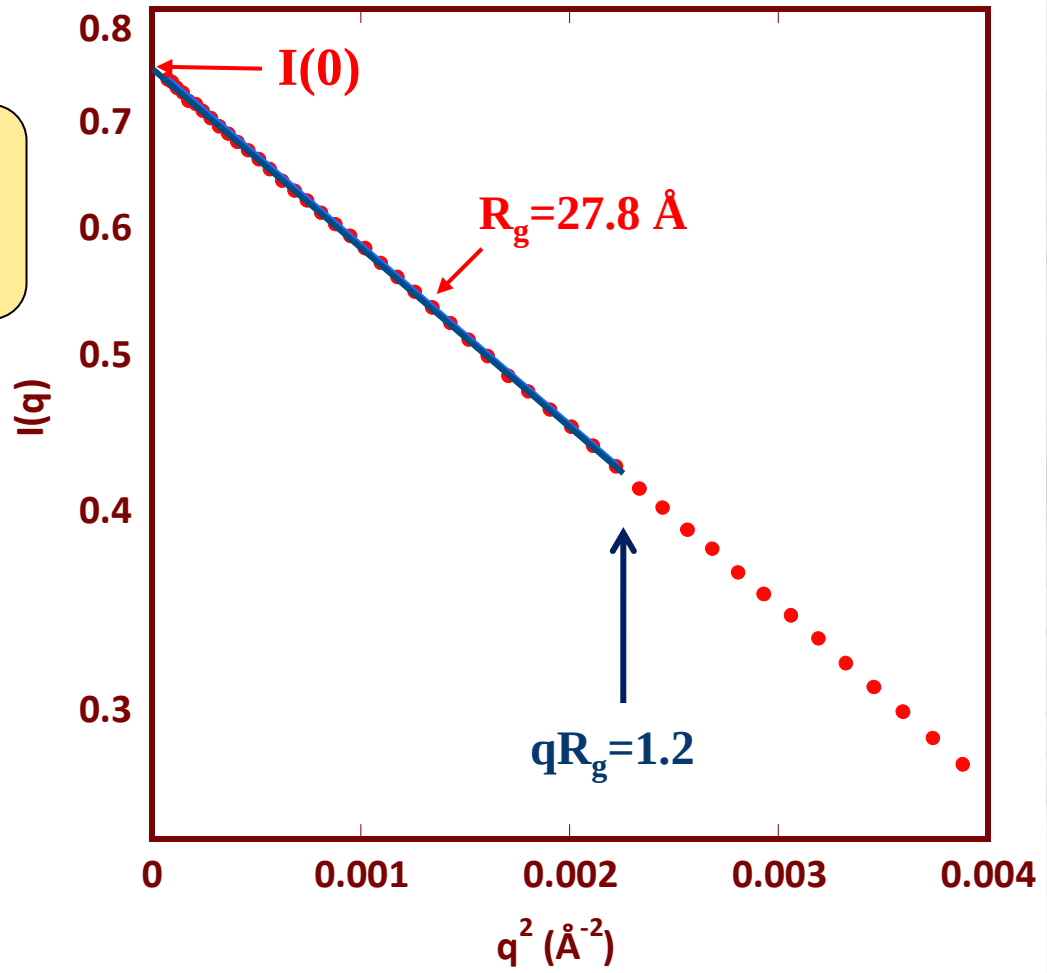
$$ln[I(q)] \cong ln[I(0)] - \frac{R_g^2}{3} q^2$$

Validity range :

$0 < qR_g < 1$ for a solid sphere

$0 < qR_g < 1.3$ rule of thumb for a globular protein

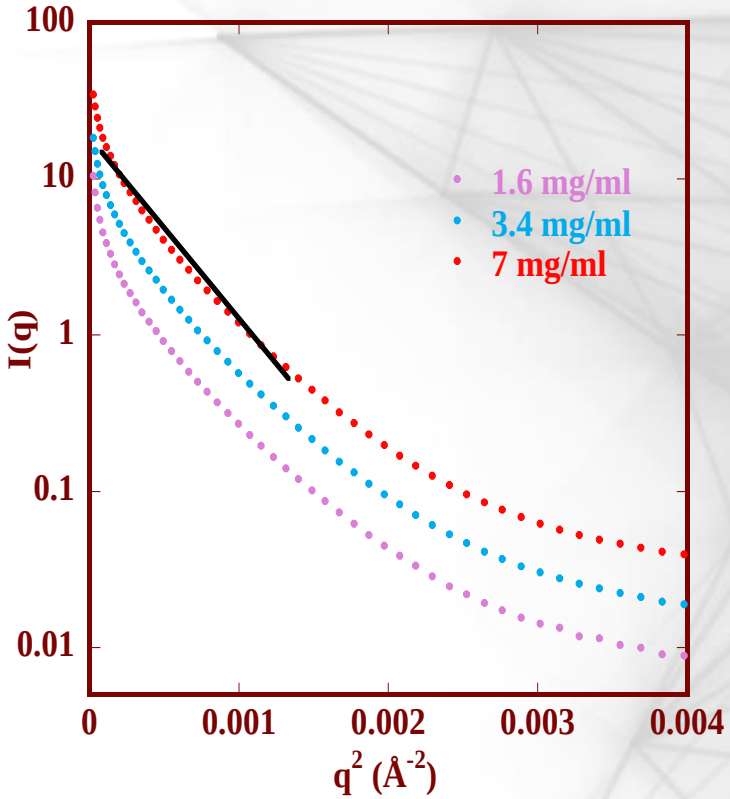
ideal monodispersed

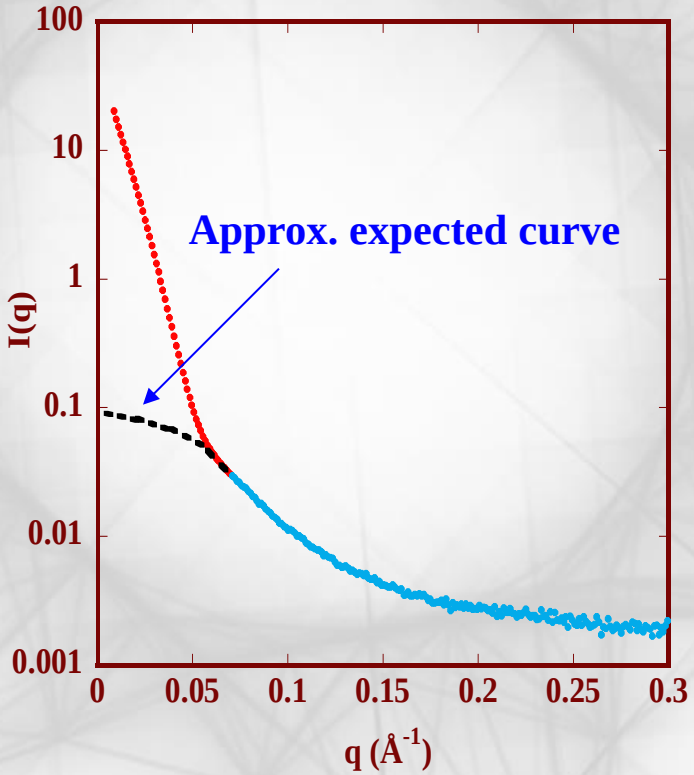


Data Analysis: Evaluation of the solution properties

Irreversible aggregation

→ Useless data: the whole curve is affected





I(0): > 150 fold the expected value for the given MM

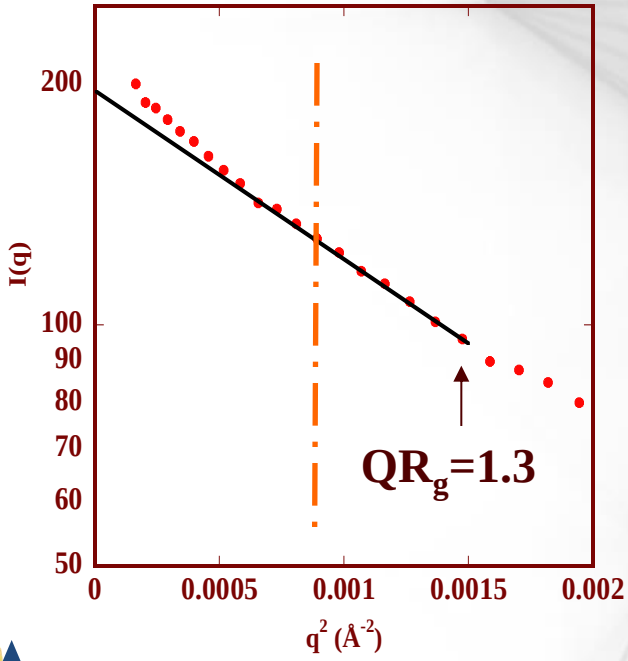
Swing – Domaine 1-242 de RRP44 – 07/08

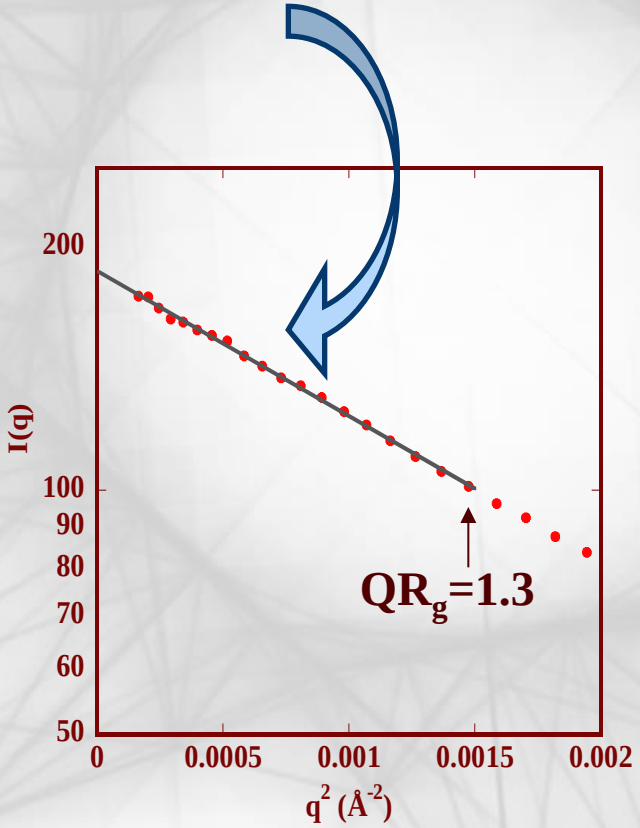
(Courtesy D. Durand, IBBMC, Orsay)

Data Analysis: Evaluation of the solution properties

Weak aggregation → **possible improvement**

centrifugation, buffer change





R_g ~ 38 Å – too high!!

R_g ~ 36 Å

Nanostar –PR65 protein
(Courtesy D. Durand, IBBMC, Orsay)

Data Analysis: Evaluation of the solution properties

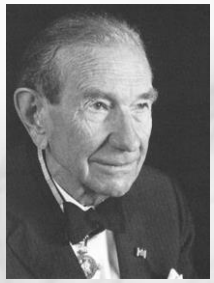
Guinier plot

- **A linear Guinier plot is a requirement, but it is NOT a sufficient condition ensuring ideality (nor monodispersity) of the sample.**

Data Analysis

- Guinier Analysis
- Kratky plot : why is it so interesting ?
- « Real-space SAXS » : Distance correlation function P(r)

Data Analysis: Kratky plot

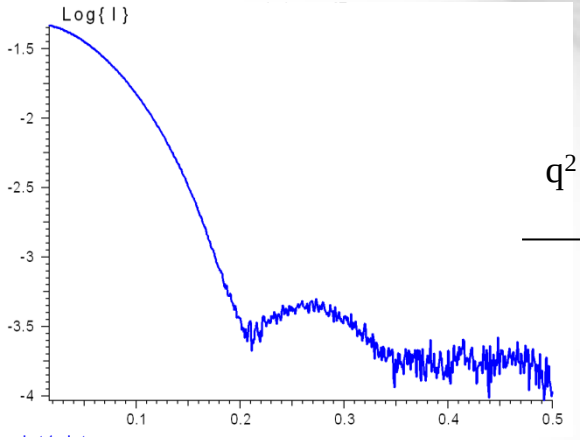


Prof. Otto Kratky
1902-1995
Graz, Austria

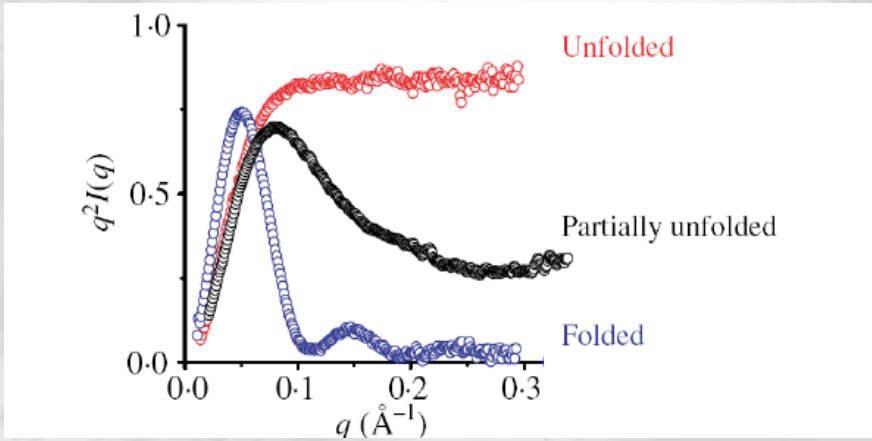
SAXS provides a sensitive means to ***evaluate the degree of compactness*** of a protein:

- To determine whether a protein is globular, extended or unfolded
- To monitor the folding or unfolding transition of a protein

This is most conveniently represented using the so-called Kratky plot:



$q^2 I(q)$ *versus* q



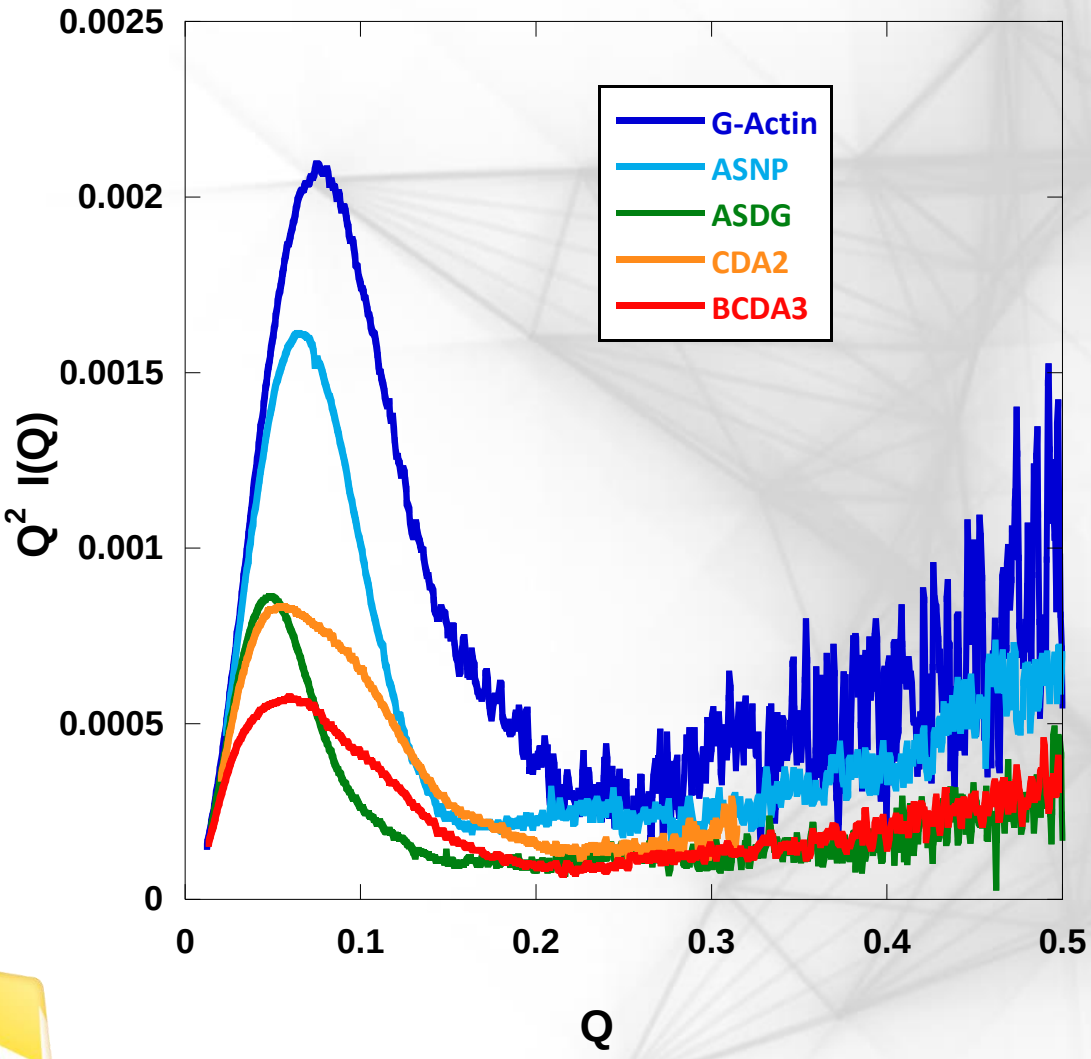
Putnam, D., et al. (2007) Quart. Rev. Biophys. 40, 191-285.

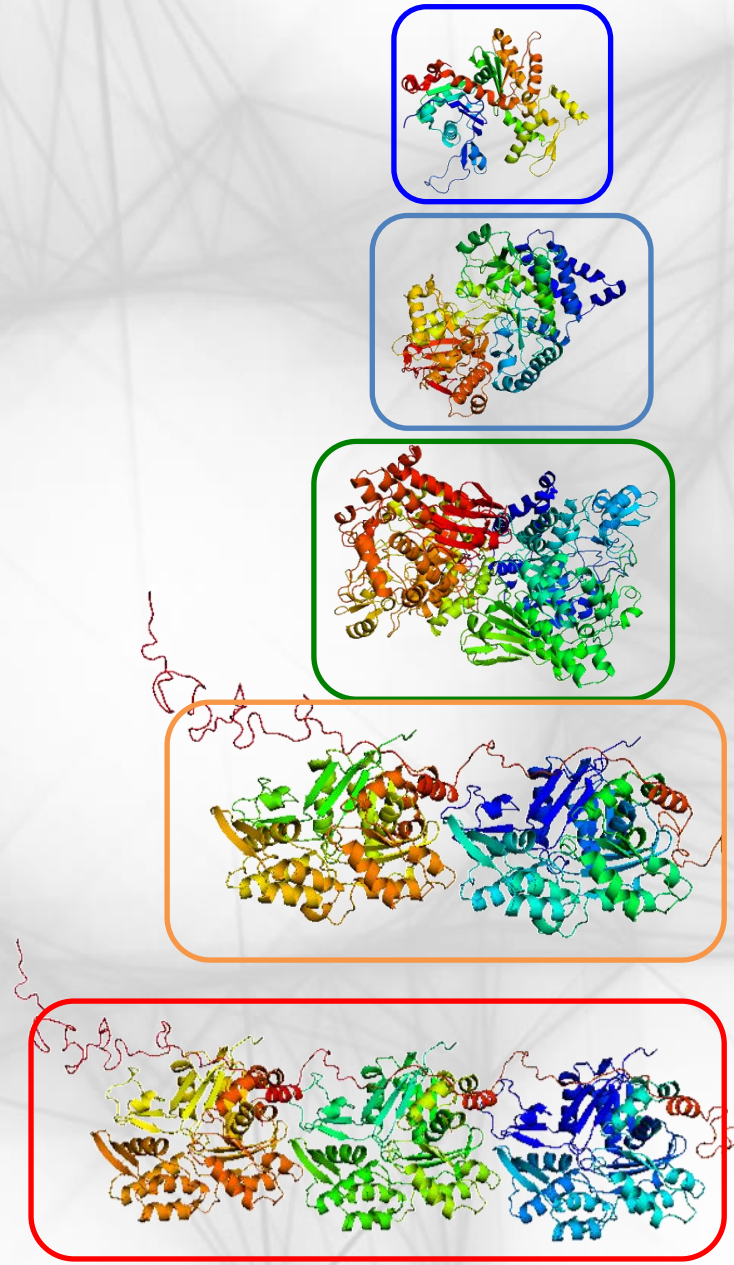
Folded particle : ***bell-shaped curve*** (asymptotic behaviour $I(Q) \sim Q^{-4}$)

Random polymer chain : ***plateau*** at large q-values (asymptotic behaviour in $I(Q) \sim Q^{-2}$)

Extended polymer chain : ***increase*** at large q-values (asymptotic behaviour in $I(Q) \sim Q^{-1.x}$)

Data Analysis: Kratky plot



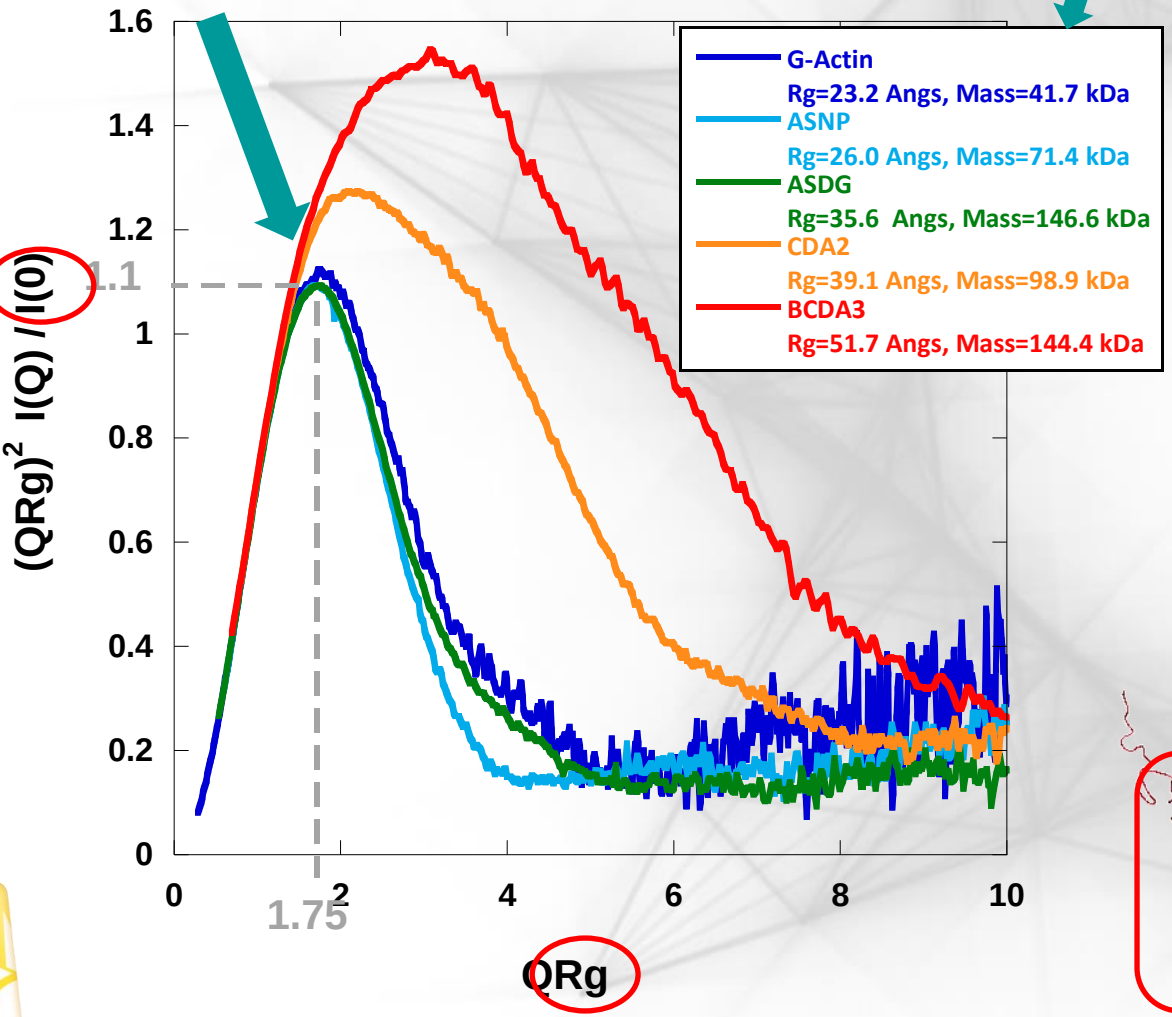


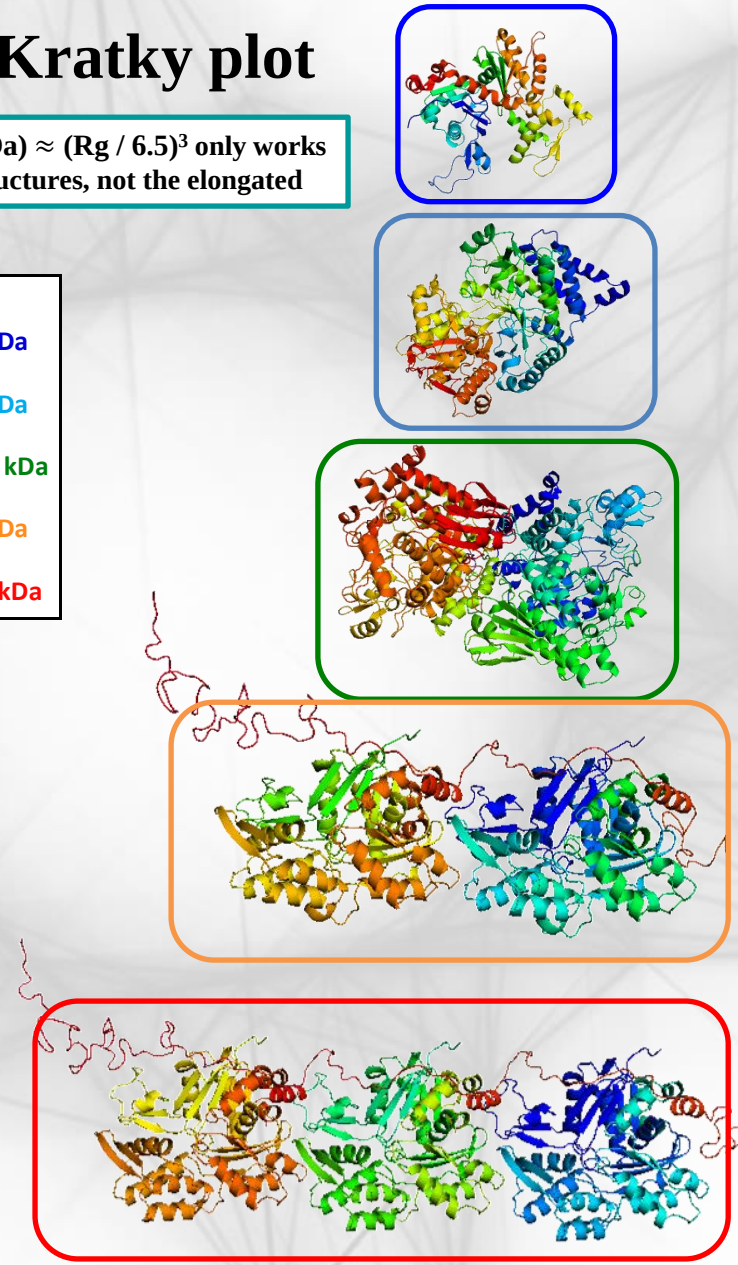
Folded proteins display a bell shape. Can we go further?

Data Analysis: Dimensionless Kratky plot

For globular structures, DLKPs fold into the same maximum

The relation $M_{Rg}(kDa) \approx (Rg / 6.5)^3$ only works for the globular structures, not the elongated

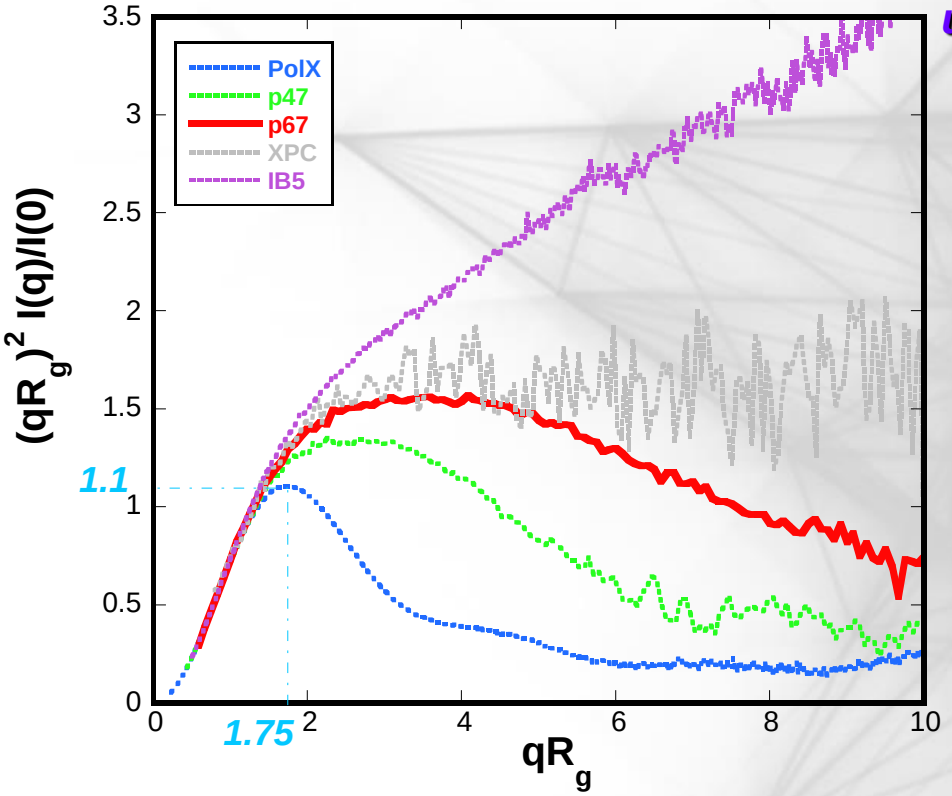




The maximum value on the dimensionless bell shape tells if the protein is globular.

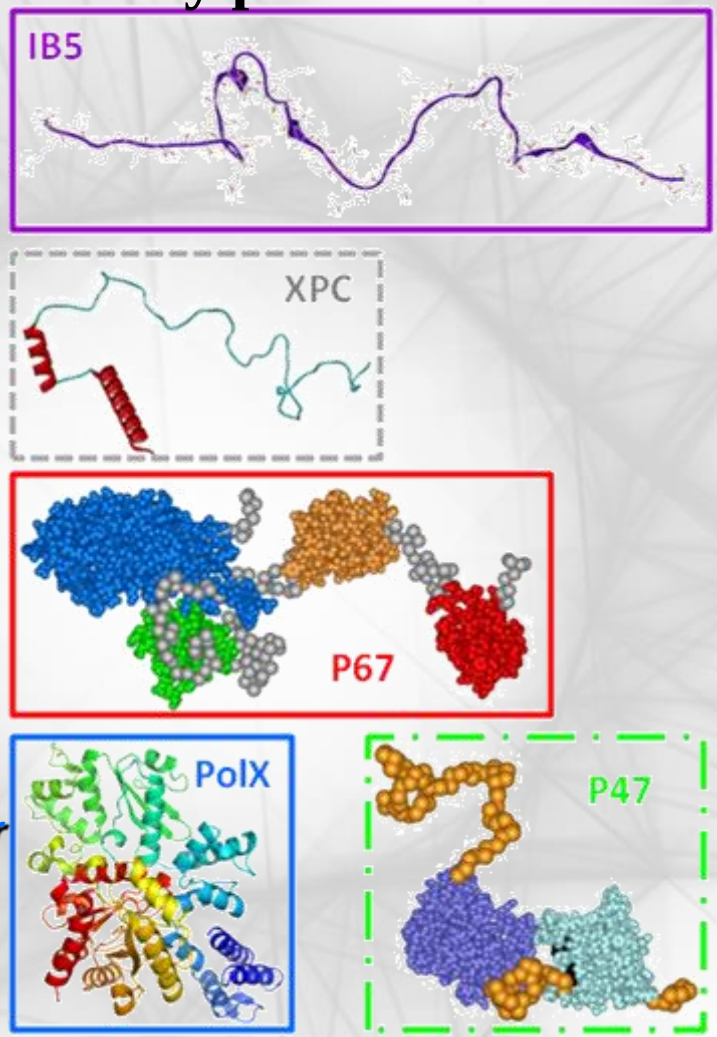
Introduced for biology in Durand et al. (2010), J. Struct. Biol. **169**, 45-53.

Data Analysis: Dimensionless Kratky plot



unfolded

globular



The bell shape vanishes as folded domains disappear and flexibility increases.

The curve increases at large Q as the structure extends.

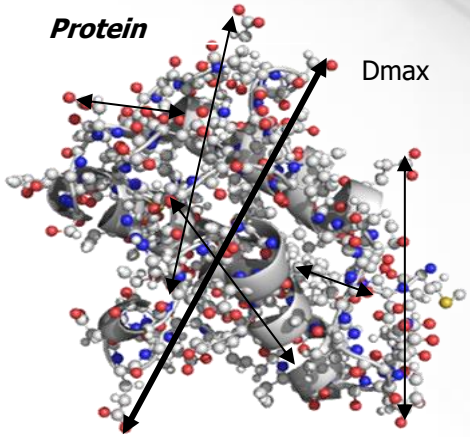
Receveur-Bréchot V. and Durand D (2012), Curr. Protein Pept. Sci., 13:55-75.

Data Analysis

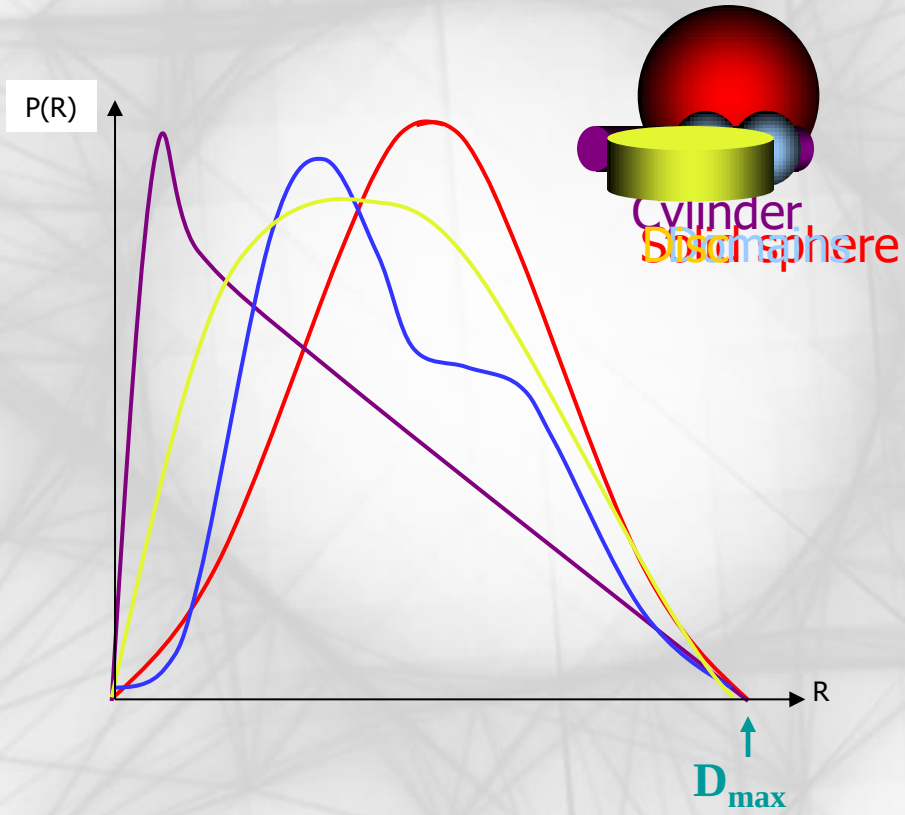
- Guinier Analysis
- Kratky plot : why is it so interesting ?
- « Real-space SAXS » : Distance correlation function P(r)

Data Analysis: Distance Distribution Function p(r)

The distance distribution function p(r) is proportional to the average number of atoms at a given distance, r, from any given atom within the macromolecule.



p(r) vanishes at r = D_{max}



The distance distribution function characterises the shape of the particle **in real space**

Data Analysis: Distance Distribution Function p(r)

Intensity is the Fourier Transform of self-correlation function $\gamma_{obj}(r)$:

$$I(q) = 4\pi r_e^2 \varphi \int_{V_{obj}} \gamma_{obj}(r) r^2 \frac{\sin(qr)}{qr} dr$$

Fourier Transform for isotropic samples

And :

$$p(r) = \gamma_{obj}(r) r^2$$

Then :

$$I(q) = 4\pi r_e^2 \varphi \int_0^D p(r) \frac{\sin(qr)}{qr} dr$$

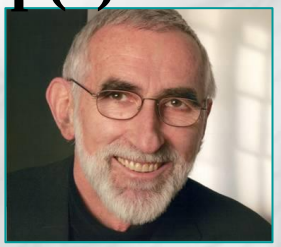
And :

$$p(r) = \frac{r^2}{2\pi^2 \varphi r_e^2} \int_0^\infty q^2 I(q) \frac{\sin(qr)}{qr} dq$$

p(r) could be directly derived from I(q). Both curves contain the same information.

However, direct calculation of p(r) from I(q) is made difficult and risky by [Qmin,Qmax] truncation and data noise effects.

Data Analysis: Distance Distribution Function p(r)



Prof. Otto Glatter
Guinier Prize 2012
Graz, Austria

Main hypothesis : the particle has a « finite » size, characterised by D_{max}.

- D_{max} is proposed by the user
- p(r) is expressed over [0, D_{Max}] by a linear combination of orthogonal functions

$$p_{theoret}(r) = \sum_{1}^{M} c_n \varphi_n(r)$$

Glatter, O. *J. Appl. Cryst.* (1977) **10**, 415-421.

- I(q) is calculated by Fourier Transform of $p_{theoret}(r)$

$$I(q) = 4\pi \, r_e^{\,2} \varphi \int_0^{D_{max}} p_{theoret}(r) \frac{\sin(q \cdot r)}{q \cdot r} dr$$

Dr. Dmitri Svergun
Hamburg, Germany

Svergun (1988) : program "GNOM"

M ~ 30 - 100 ⇒ ill-posed LSQ ⇒ regularisation method

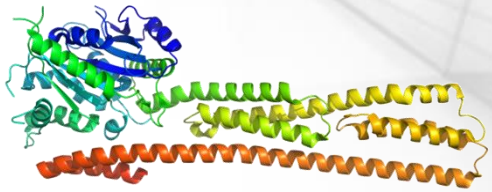
+ "Perceptual criteria" : smoothness, stability, absence of systematic deviations

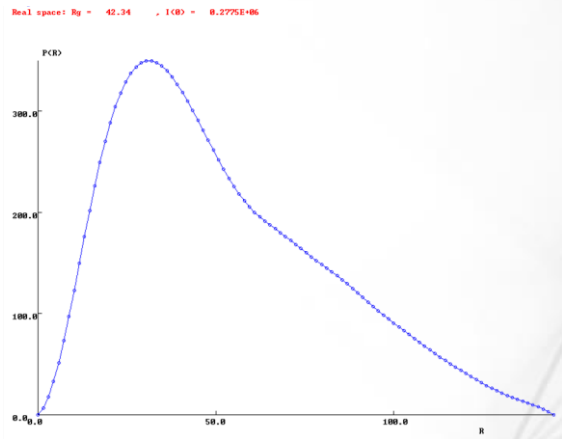
- Each criterium has a predefined weight
- The solution is given a score calculated by comparison with « ideal values »

Data Analysis: Distance Distribution Function p(r)

Experimental examples

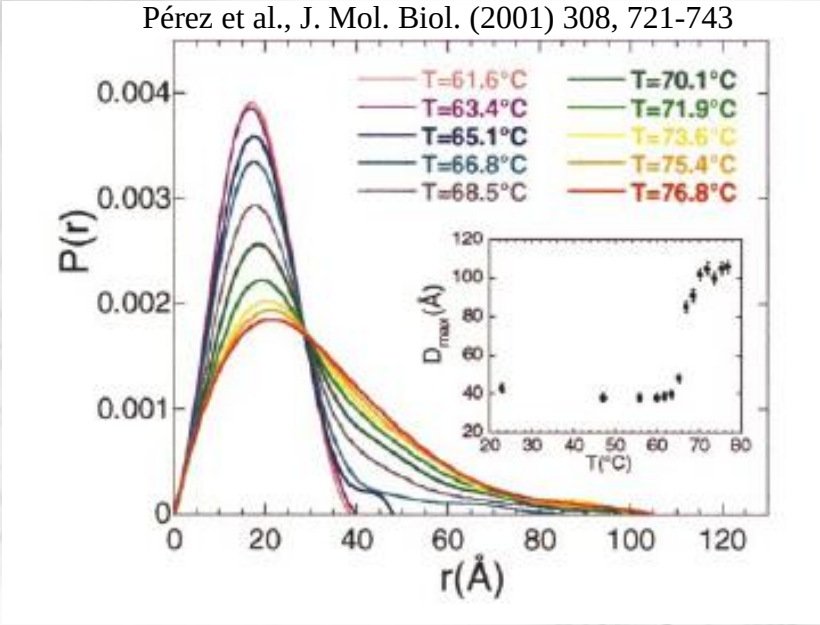
GBP1





Heat denaturation of Neocarzinostatin

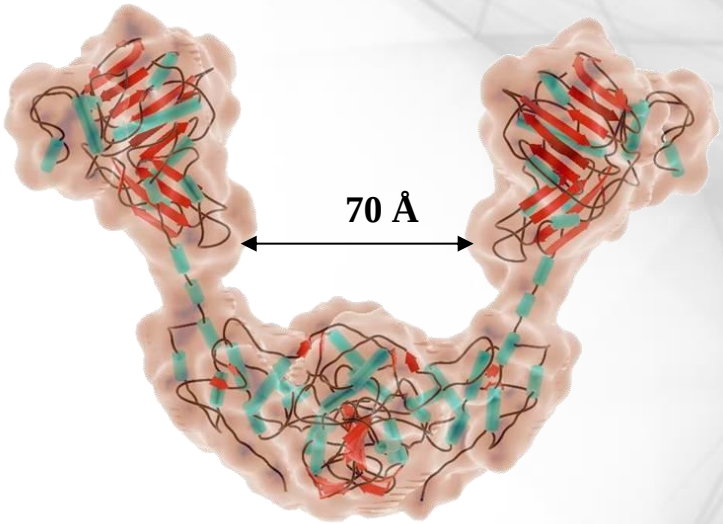
Pérez et al., J. Mol. Biol. (2001) 308, 721-743



Data Analysis: Distance Distribution Function p(r)

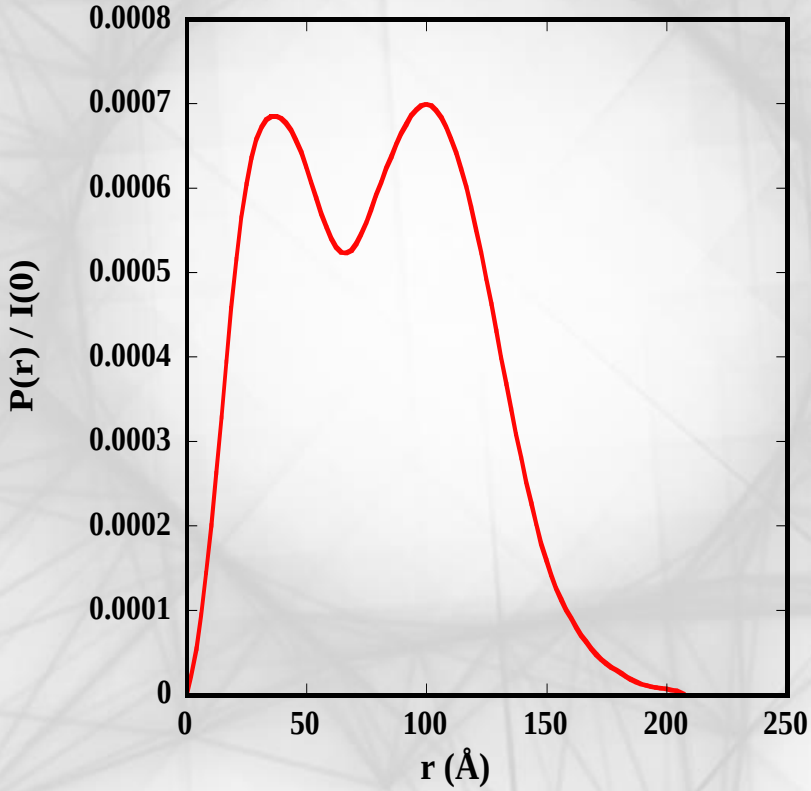
Experimental examples

Topoisomerase VI



M. Graille et al., Structure (2008), *16*, 360-370.

Bimodal distribution



Data Analysis: strategy

Data analysis

Guinier approximation
- Rg (size) and I(0) (mass and oligomeric state)

Kratky plot
- type of structure (globular, elongated or unfolded)

Distance distribution function p(r) :
- Dmax evaluation
- Rg (size) and I(0) compatibility with Guinier approximation
- Global form of the object

Molecular modeling

Nothing is known
- low resolution shape

Cristallographic , NMR structures or complete molecular modeling
- theorical curves calculation and data comparison

Structures of subunits available
- molecular modeling rigid body against SAXS data

Structures with missing loop or flexible parts
- molecular modeling of missing parts against SAXS data

Molecular Modeling: strategy

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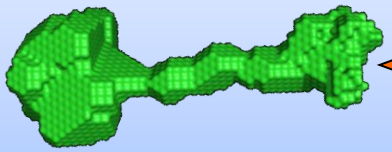
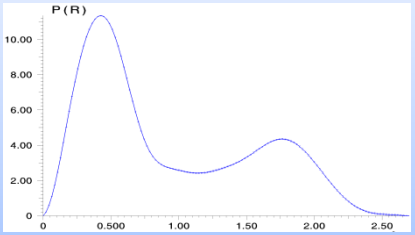
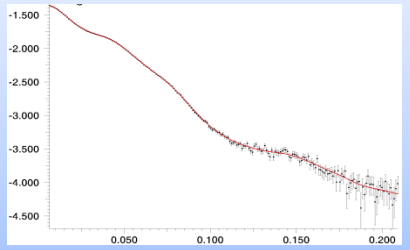
Molecular Modeling: strategy

Nothing known (except the curve)

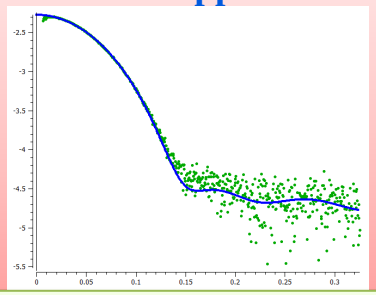


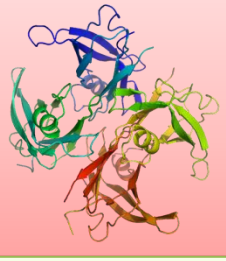


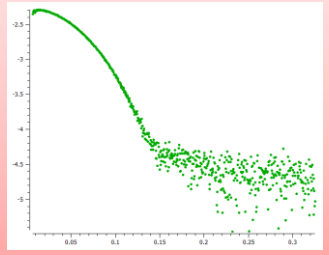


DAMMIN
DAMMIF
DENFERT

Known or supposed all-atom models

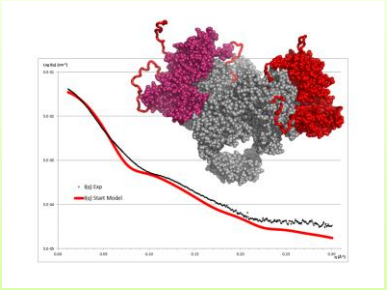






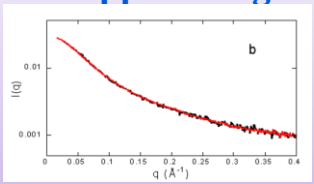
CRYSOL
FOXS
WAXIS
PEPSI-SAXS

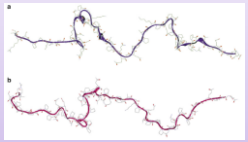
atomic structures of domains are known



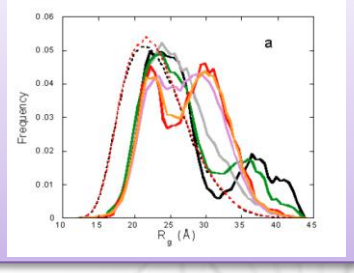
SASREF
BUNCH
CORAL
DADIMODO

Zones of supposed high flexibility





Selection within an Ensemble of Random Conformations



EOM

Molecular Modeling: Common features to all approaches

- ✓ *Monte-Carlo based methods (simulated annealing, genetic algorithm) : no unique solution.*

  → *repeat the calculation ca 10 times.*

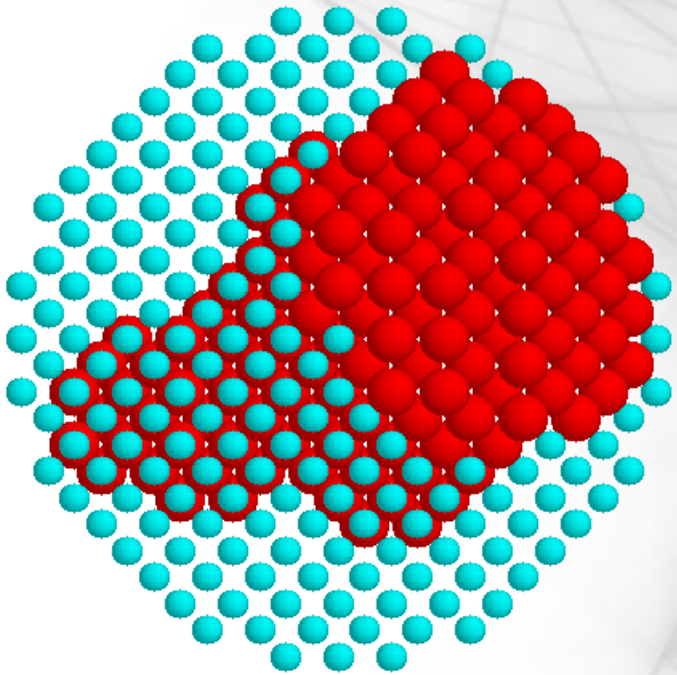
  → *repeat the calculation n x 100 times followed by clustering.*

- ✓ *make use of constraints to restrict the solution space to (bio)physically meaningful models. The program minimizes the sum of the χ^2 with experimental data and penalty terms such as:*

$$f(X) = \sum_i \chi_i^2 + \alpha_{dist} P_{dist}(X) + \beta_{cross} P_{cross}(X) + \gamma_{cont} P_{cont}(X)$$

Molecular Modeling: Nothing known (except the curve)

Initial volume : sphere diameter D_{max}



Position(j) = X(j) = 1 or 0

- $M \approx (D_{max}/r_0)^3 \approx 10^3 >> N_s$ parameters, too many for conventional minimization
- No unique shape restoration unless constrained
- Able to describe complex shapes

Slide from Dmitri Svergun, EMBL Hamburg

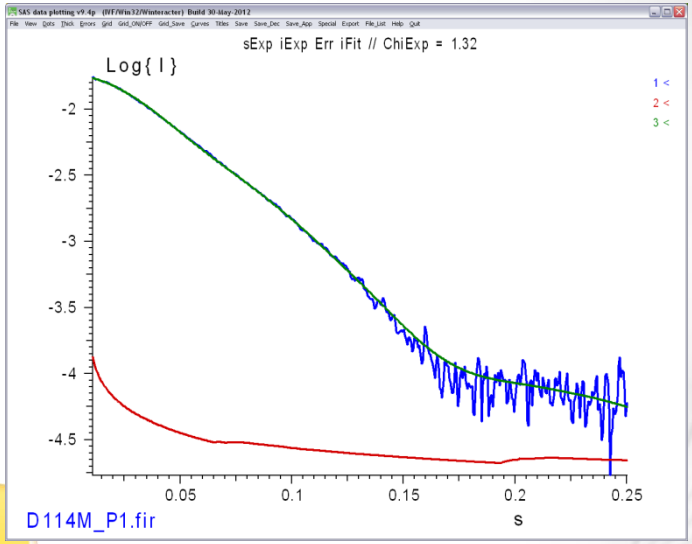
Chacón, P. *et al.* (1998) *Biophys. J.* **74,** 2760-2775.

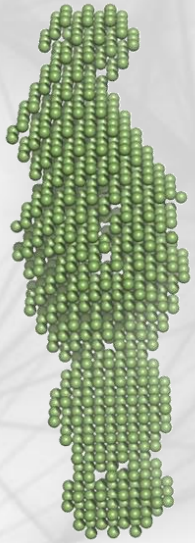
Walter, D., Cohen, F.E. & Doniach, S. (1999), J. Appl. Cryst.,33, 350-363

Molecular Modeling: Nothing known (except the curve)

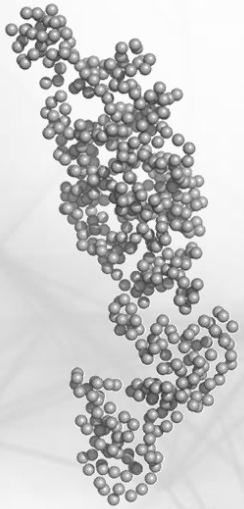
DAMMIN/DAMMIF : very low resolution, restricted portion of the data used ($q < 0.2$ Å^{-1}), very basic constraints

GASBOR : a protein comprising N residues is represented by an ensemble of N spheres centered at the Cα positions, the whole q-range can be used.
An initial gas-like distribution of dummy residues is refined using Simulated Anneling to fit the data under constraints ensuring a final chain like distribution

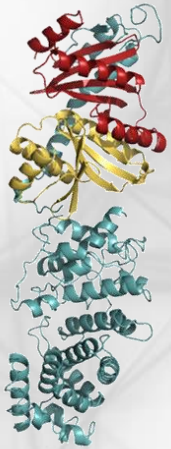




DAMMIF shape



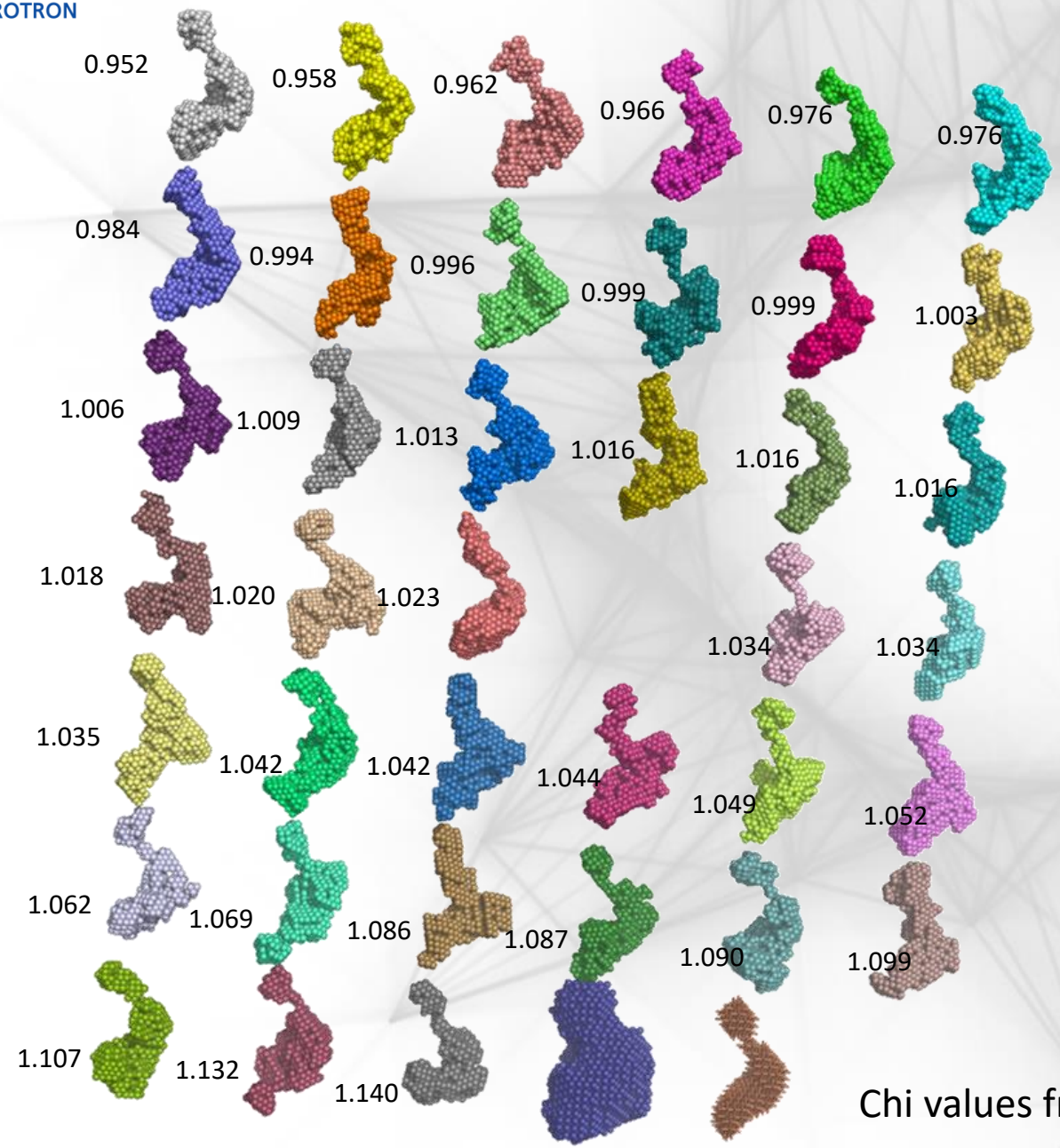
GASBOR beads model



High resolution structure

D. Svergun et al.(2001), Biophys. J., 80, 2946-2953.

Molecular Modeling: Nothing known (except the curve)

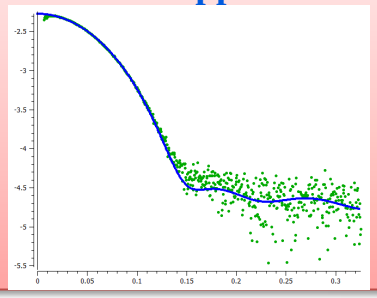


NSD values

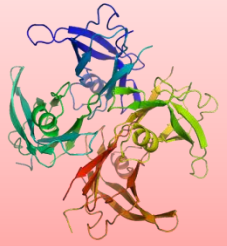
Chi values from 1.704 to 1.942

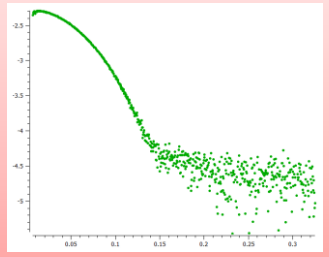
Molecular Modeling:

Known or supposed all-atom models



validation / elimination

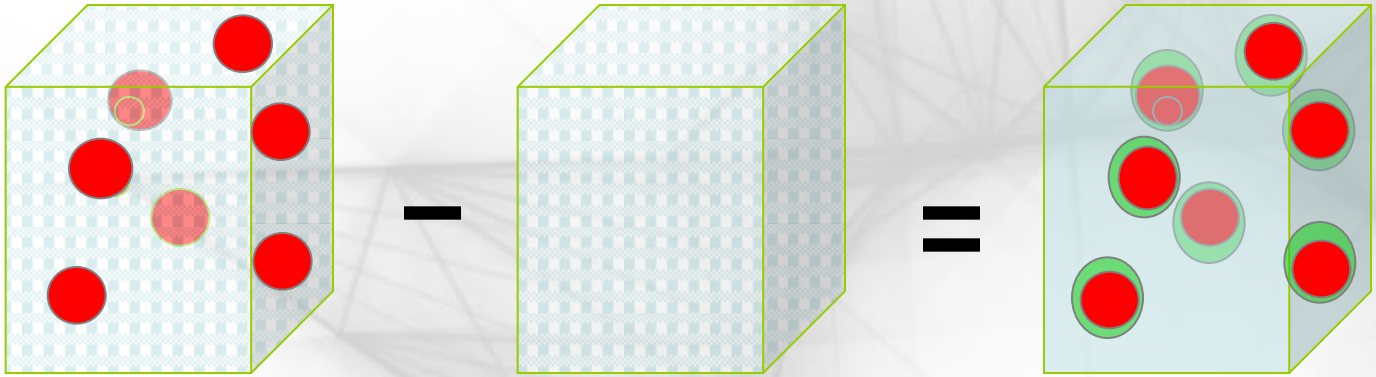




CRYSOL
FOXS
WAXIS
PEPSI-SAXS

From a atomic structure to a solution scattering pattern : program CRYSOL

Molecular Modeling: Solvent scattering and contrast



$I_{solution}(q)$ $I_{solvent}(q)$ $I_{particle}(q)$

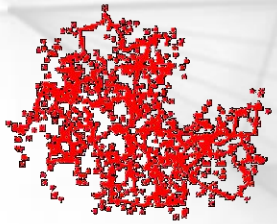
The bound solvent density differs from that of the bulk.

Bulk water density = 0.334 $e^-/Å^3$

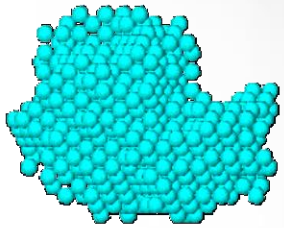
Hydration layer density ~ 5-15 % higher

Molecular Modeling: Scattering from a macromolecule

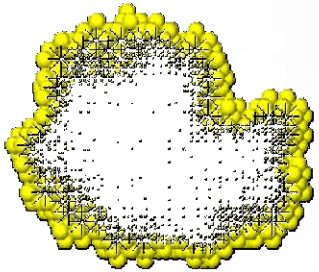
$$I(s) = \left\langle \left| A(\mathbf{s}) \right|^2 \right\rangle_\Omega = \left\langle \left| A_a(\mathbf{s}) - \rho_s A_s(\mathbf{s}) + \delta\rho_b A_b(\mathbf{s}) \right|^2 \right\rangle_\Omega$$



- $A_a(s)$: atomic scattering in vacuum



- $A_s(s)$: scattering from the excluded volume



- $A_b(s)$: scattering from the hydration shell, layer of thickness 3 Å

CRYSOL (X-rays): Svergun et al. (1995). *J. Appl. Cryst.* 28, 768
CRYSON (*neutrons):* Svergun et al. (1998) *P.N.A.S. USA*, 95, 2267

Molecular Modeling: Scattering from a macromolecule

Svergun D, Barberato C, and Koch M.H.J. (1995) **CRYSOL** – a program to evaluate x-ray solution scattering of biological macromolecules from atomic coordinates.
J. Appl. Cryst. 28, 768
Most popular for BioSAXS, stand-alone program, fit model to data, fast computational algorithm . ***1500 citations***.
http://www.embl-hamburg.de/biosaxs/atsas-online/crysol.php

Grishaev A, Guo L, Irving T, Bax A. (2010) **AXES** Improved Fitting of Solution X-ray Scattering Data to Macromolecular Structures and Structural Ensembles by Explicit Water Modeling. J. Am. Chem. Soc. 132**,** 15484-6.
Use explicit water modeling solvation layer, robust fitting approach
http://spin.niddk.nih.gov/bax/nmrserver/saxs1/

Schneidman-Duhovny D, Hammel M, Sali A. (2010) **FoXS**: a web server for rapid computation and fitting of SAXS profiles. Nucleic Acids Res. 3**8** Suppl:W540-4.
Debye-like computation, web server based. Hydration taken into account by "inflating" the volume of surface atoms.
http://modbase.compbio.ucsf.edu/foxs/

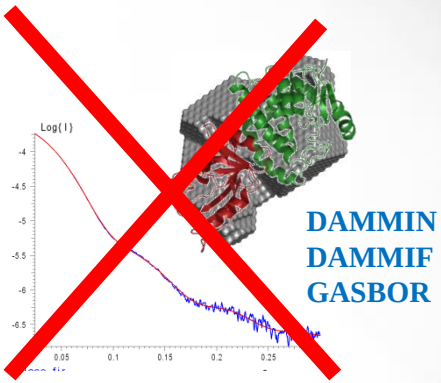
Knight C. J. and S. Hub J. S. (2015) **WAXSiS**: a web server for the calculation of SAXS/WAXS curves based on explicit-solvent molecular dynamics.
Nucleic Acids Res. 43 Suppl: W225-30.
http://waxsis.uni-goettingen.de

Grudinin S, Garkavenko M, Kazennov A. (2017) **Pepsi-SAXS:** an adaptive method for rapid and accurate computation of small-angle X-ray scattering profiles. Acta Crystallogr D Struct Biol.
https://team.inria.fr/nano-d/software/pepsi-saxs/

Molecular Modeling: A word of caution, what NOT to do

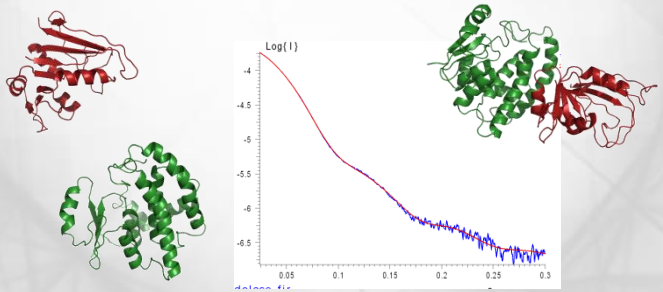
- **Common misconception: dummy atom ab initio envelope from DAMMIF (or from Gasbor for that matter) are viewed as similar to EM density maps: NO.**

- **One should not try and superimpose 3D models of domains in the envelope. There is not 1 but MANY similar (or not) envelopes. One must try and refine the position of domains vs SAXS data.**



DAMMIN
DAMMIF
GASBOR

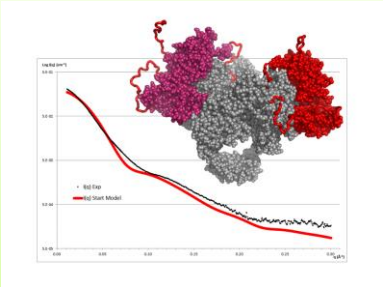
SASREF



- **Furthermore, in some cases, the volume or envelope notion is simply irrelevant: for instance, for flexible multi domain proteins or even worse, for a flexible IDP.**

Molecular Modeling: structures of domains are known

atomic structures of domains are known



SASREF
BUNCH
CORAL
DADIMODO

When atomic structures of domains are known, but not their mutual arrangement

Molecular Modeling: structures of domains are known

SASREF : when atomic structures of domains are known, but no their mutual organization

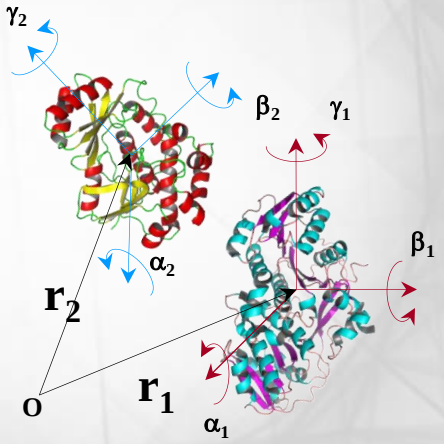
The objective is to find the relative orientation and position of each subunit that gives a good agreement with the SAXS data of the complex.

The scattering intensity I(q) of the complex is equal to the sum squared of the amplitudes of all subunits

$$I(q) = \left\langle \left| \sum_{k=1}^{K} A^{(k)}(\vec{q}) \right|^2 \right\rangle_{\Omega}$$

$$A^{(k)}(\vec{q}) = \exp(i.\vec{q}.\vec{r}_k^{\rho}) \prod (\alpha_k.\beta_k.\gamma_k)[C^{(k)}(\vec{q})]$$

Amplitudes are calculated with CRYSOL from the high resolution structure of each subunit.



The algorithm of minimization is the same used with DAMMIN with a penalty function (interconnectivity of the subunits, the steric clashes) and possibility to give information about contacting residues from other experiences.

$$f(X) = \sum_i \chi_i^2 + \alpha_{dist} P_{dist}(X) + \beta_{cross} P_{cross}(X) + \gamma_{cont} P_{cont}(X)$$

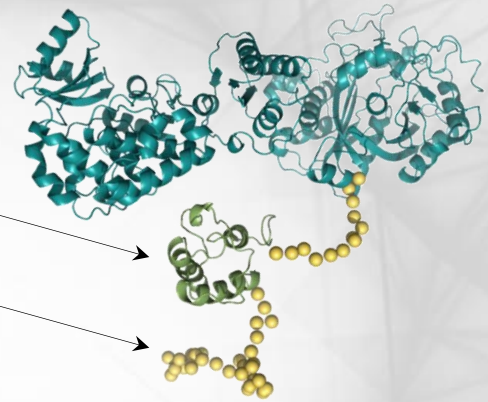
Petoukhov & Svergun (2005). Biophys. J., 89, 1237-1250.

Molecular Modeling: structures of domains are known

BUNCH and CORAL : *quaternary structure analysis of multidomain protein*

Combination of rigid body and ab initio modeling :

- *position and orientation of rigid domains*
- *possible conformation of flexible linkers*



$$f(X) = \sum_i \chi_i^2 + \alpha_{ang} P_{ang}(X) + \beta_{cross} P_{cross}(X) + \gamma_{dih} P_{dih}(X) + \delta_{ext} P_{ext}$$

As SASREF, the amplitude are calculated with CRYSOL from the high resolution structure of each monomer

The algorithm of minimization is the same used with SASREF with a penalty function including the steric clashes Pcross, the dihedral angle Pang and Pdih, and the compactness of the loop Pext. The possibility to give information about contacting residues from other experiences is also added.

Flexibility → no unique structure !
NOT a structure but a SAXS data compatible model

Petoukhov & Svergun (2005). Biophys. J., 89, 1237-1250.

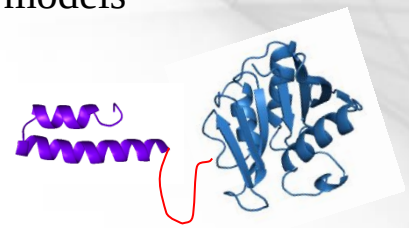
Molecular Modeling: structures of domains are known

DADIMODO : rigid body refinement *vs.* SAXS / NMR data

Modelling approach : complete atomic model

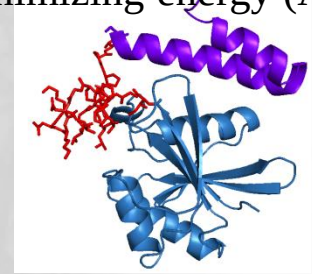
Full structure initiated with :

- Crystal or NMR domain structures
- Homology models



External information:

- Sequence
- Sub-parts moved as rigid-bodies (user-defined)
- A correct stereochemistry is maintained at all steps by minimizing energy (Amber 99 Force Field)

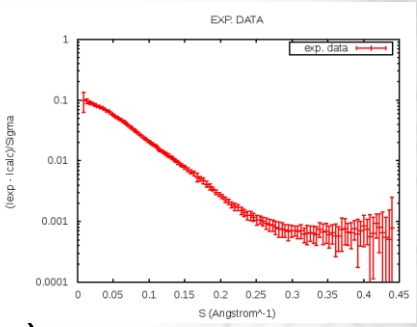


Experimental data:

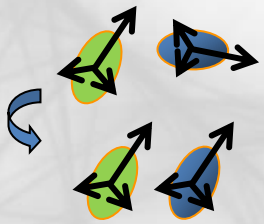
- SAXS
- NMR

  RDC

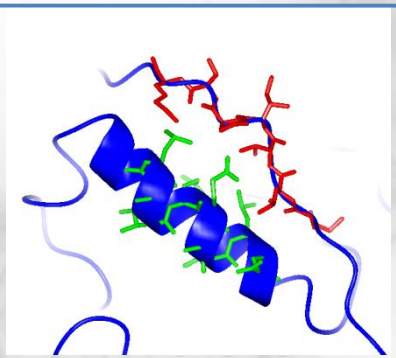
  ADR (chem. shift map.)



SAXS score



RDC score



ADR score

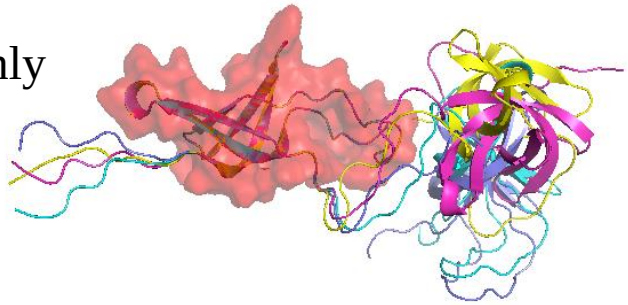
Optimisation of the structure via a genetic algorithm

Collab : F. Mareuil, C. Sizun & F. Bontems (ICSN, Gif sur Yvette))

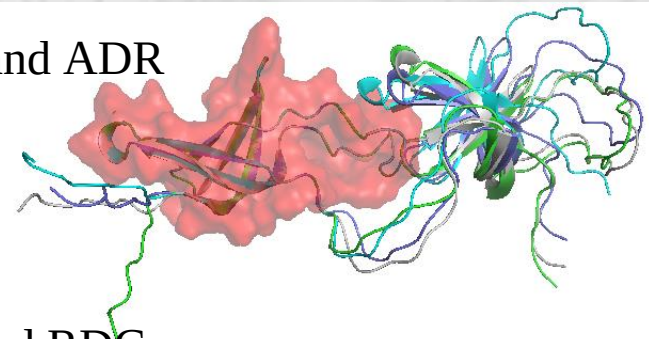
Evrard et al. (2011), *J. Appl. Cryst.*, **44**:1264-1271.

Molecular Modeling: structures of domains are known

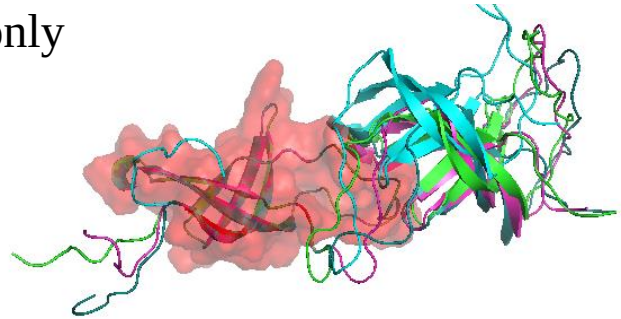
vs. SAXS only



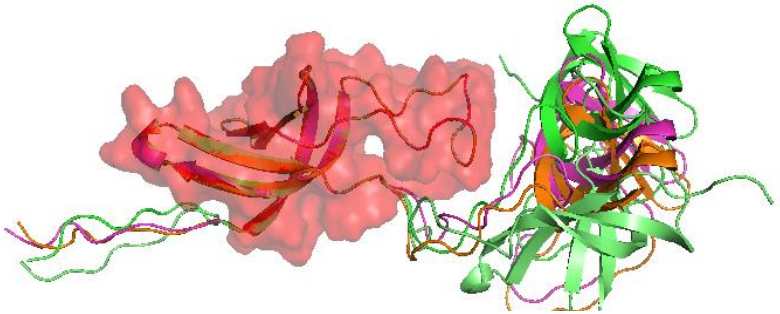
vs. SAXS and ADR



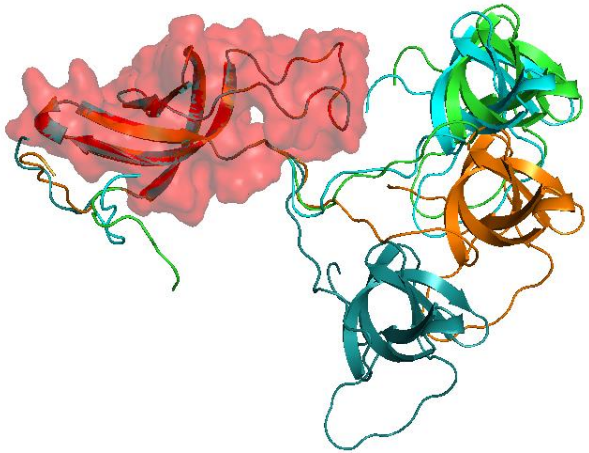
vs. ADR only



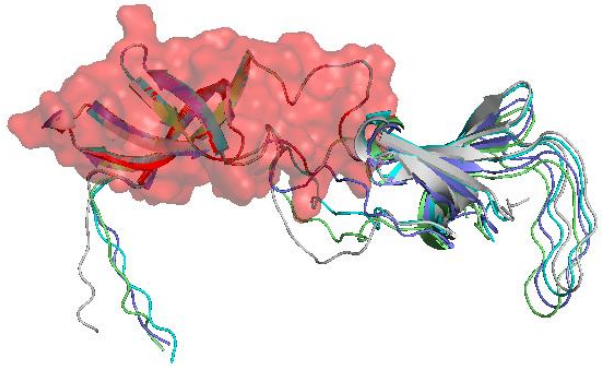
vs. SAXS and RDC



vs. RDC only



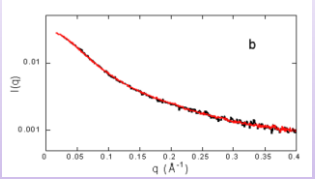
vs. SAXS, ADR, RDC

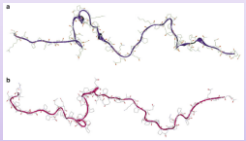


https://dadimodo.synchrotron-soleil.fr

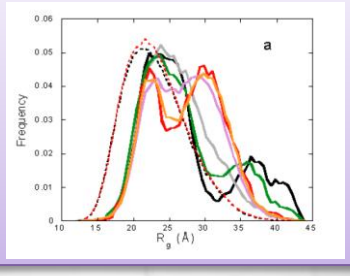
Molecular Modeling: Zones of supposed high flexibility

Zones of supposed high flexibility



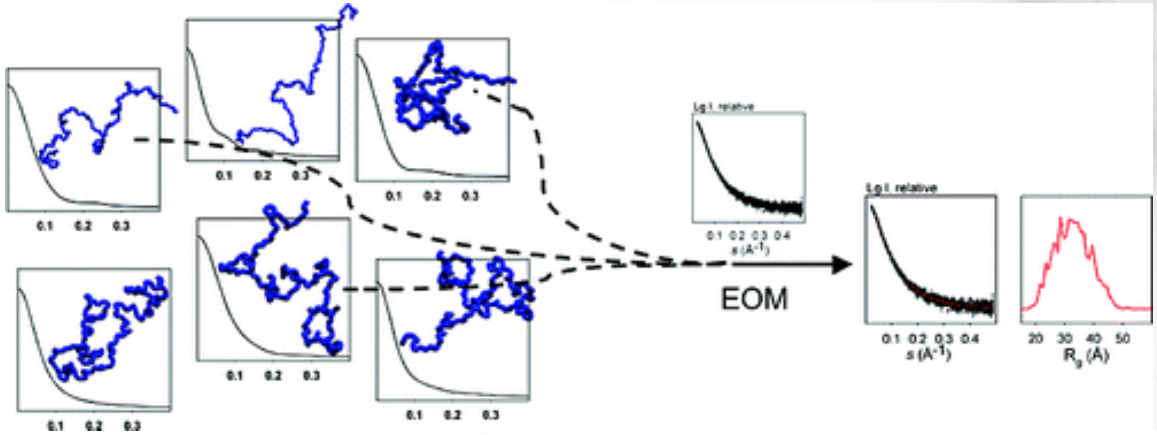


Selection within an Ensemble of Random Conformations



EOM

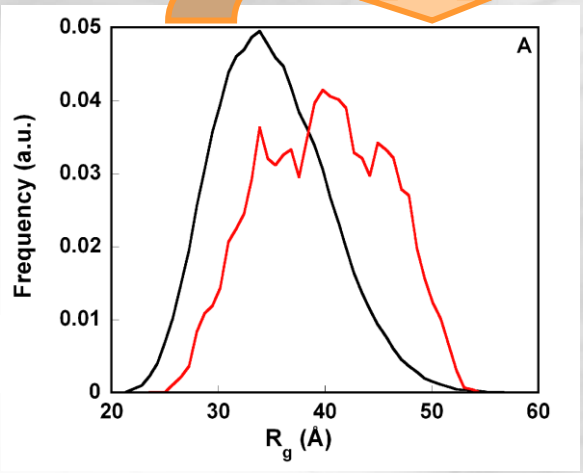
Molecular Modeling: Zones of supposed high flexibility



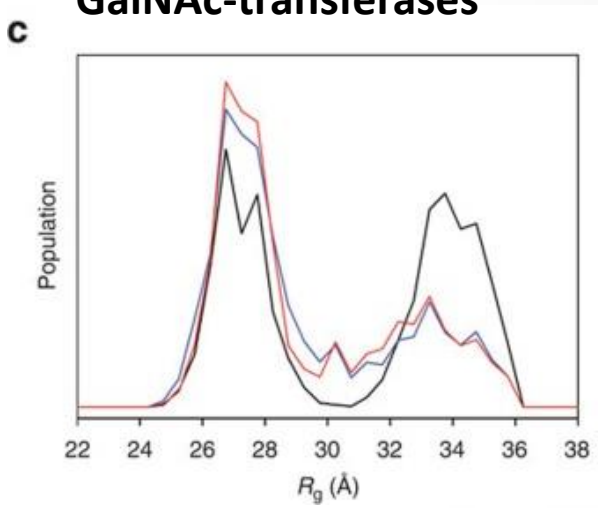
pool **optimized ensembles**

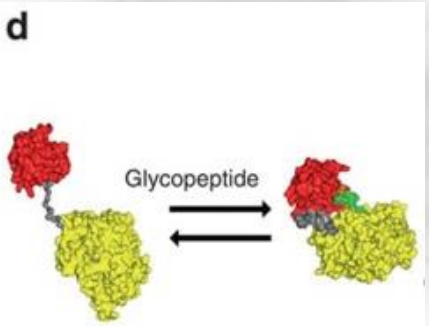
Bernadó et al, J. Am. Chem. Soc 2007

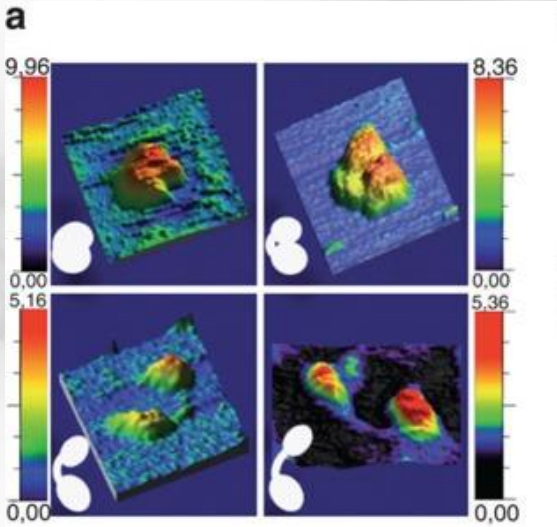
R_g distribution



GalNAc-transferases





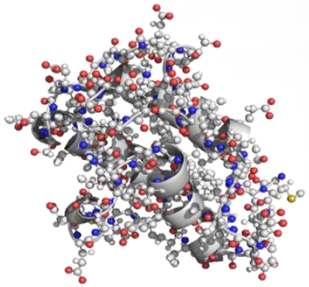


Lira-Navarette et al, Nat. Commun. 2015

Molecular Modeling: SAXS for 3D structure reconstitution

The 1D SAXS profile is the Fourier transform of the p(r) function. Contrary to direct scattering calculation, the inverse problem cannot be solved analytically, i.e., no "inverse computation" can be used to yield 3D position coordinates from scattering data.

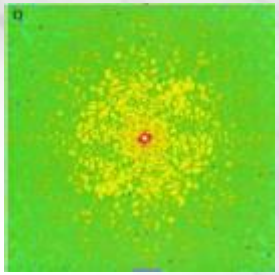
Real space 3D Molecule



Fixed orientation

Phase lost

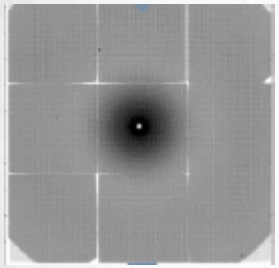
Reciprocal space
2D anisotropic image



Averaged orientation

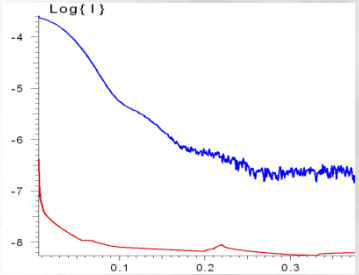
Orientation lost

Reciprocal space
2D isotropic image



Radial averaging

1D profile reciprocal space



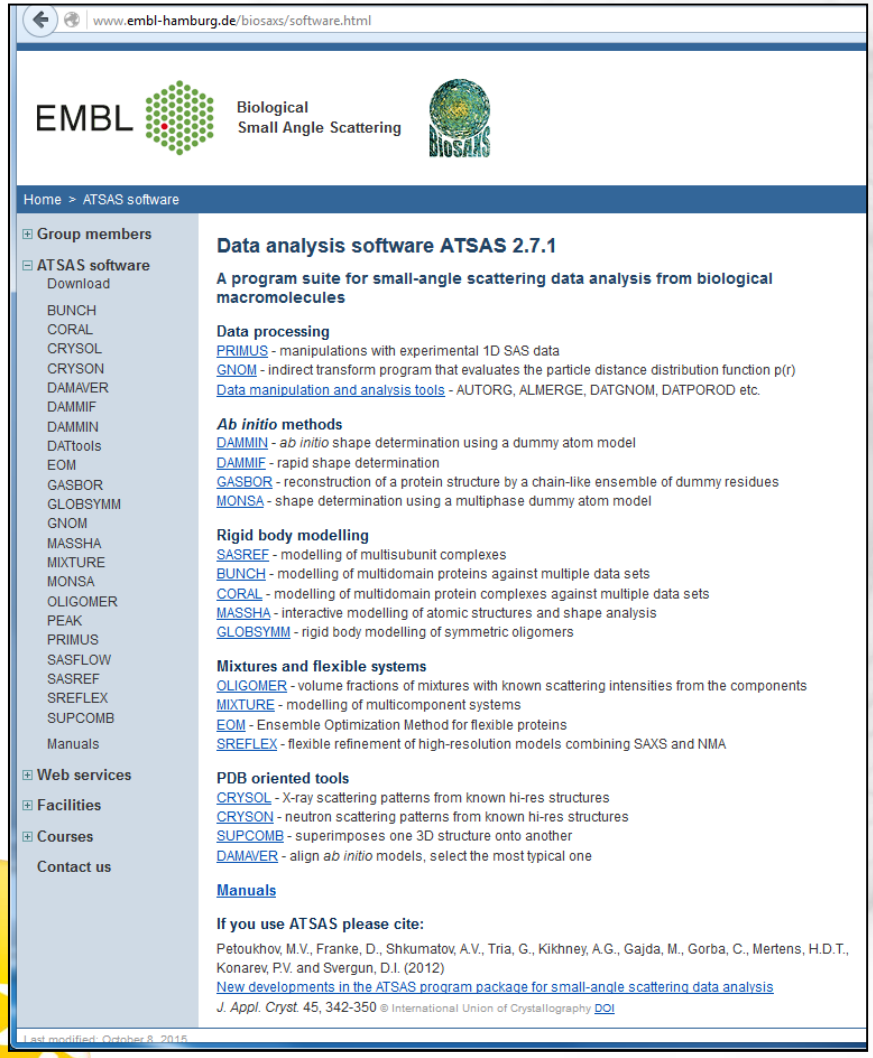
How to reconstruct the 3D structure from the 1D SAXS profile ?

Bear in mind !

One 3D structure → One SAXS curve
BUT
One SAXS curve → Many 3D structures, all compatible with the same curve
Additional constraints are always needed

ATSAS package and ATSAS online

http://www.embl-hamburg.de/biosaxs/software.html



http://www.embl-hamburg.de/biosaxs/atsas-online/

CONCLUSION

- **A scattering pattern can be calculated from atomic coordinates, thereby providing a link between crystal and solution work.**

- **Using SAXS patterns, a*b initio* methods can determine the shape of a molecule**

- **Rigid-body modeling allows one to propose models for complexes best fitting the data.**

- **Useful though limited structural information about flexible systems can be derived from SAXS data.**

COMMENTS

- ✓ SAXS is at his best when it is used to distinguish between several preconceived hypotheses.
- ✓ Analysis and modeling require a monodisperse and ideal solution, which has to be checked independently.
- ✓ Otherwise :

IN

SAXS

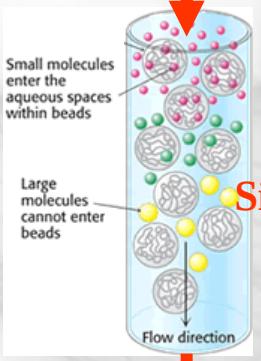
OUT

FEW EXPERIMENTAL CONSIDERATIONS

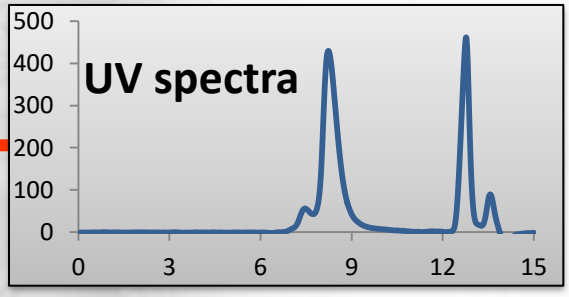
AutoSampler / SEC-SAXS

Flow rate 300 µl/min

- Monodisperse solution
- Aggregation is eliminated
- Oligomeric conformations can be distinguished
- Equilibrium states can be transiently separated
- Perfect background subtraction
- Automatic concentration series



Size Exclusion



Incident X-ray

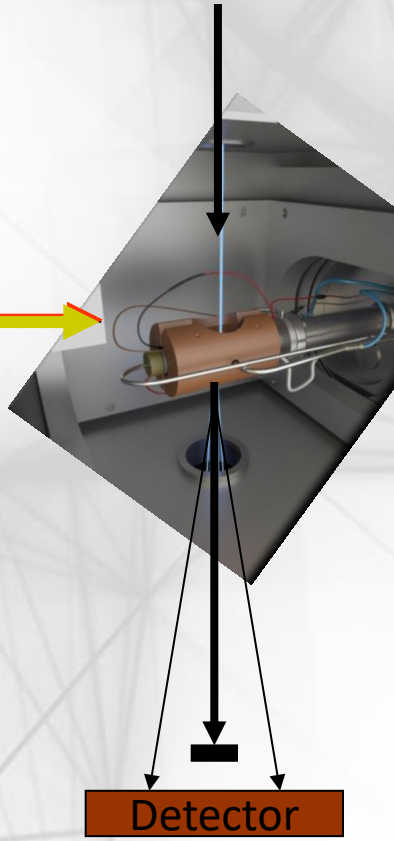
Samples injection:

- Sample [C4]

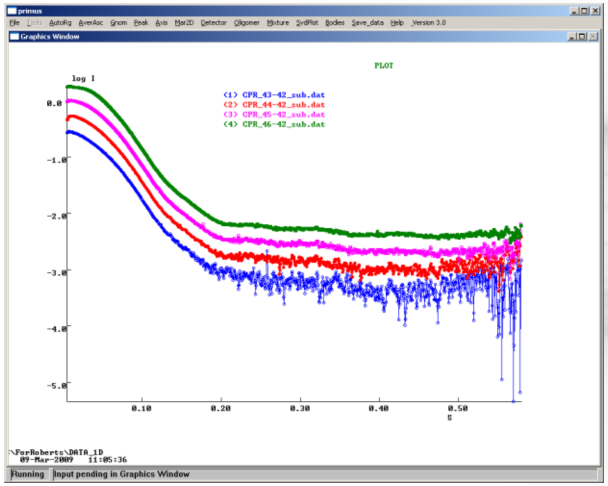
Flow rate 75 µl/min

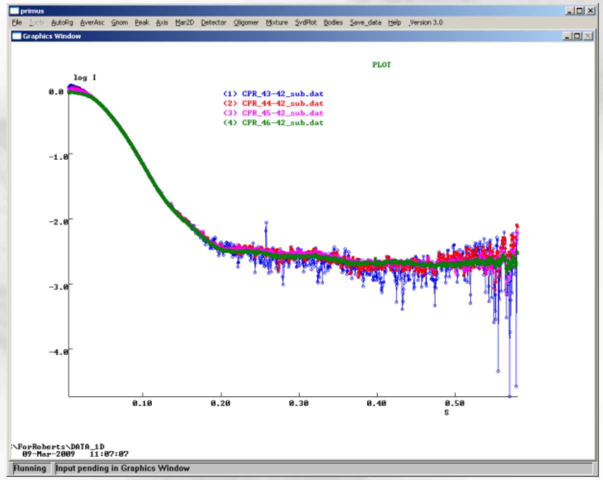
Pure sample

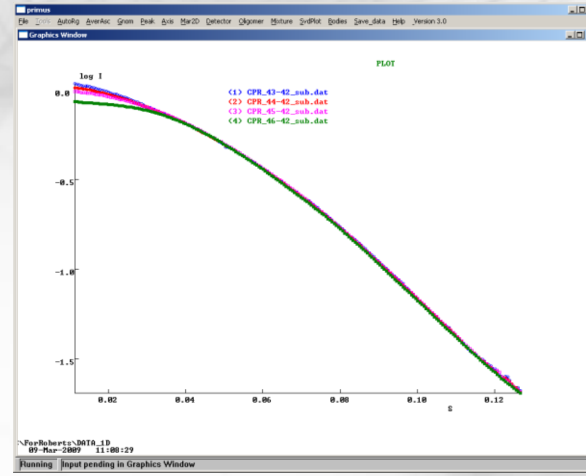
- Small volumes (~ 10 to 40 µl)
- No dilution
- High rate (~2 minutes/sample)
- Check ideality

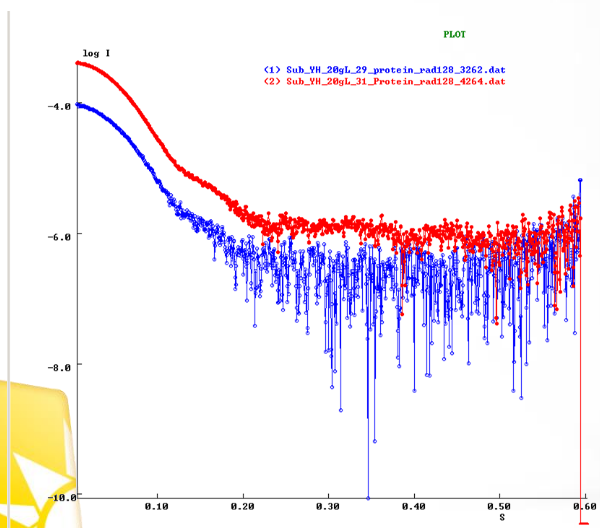


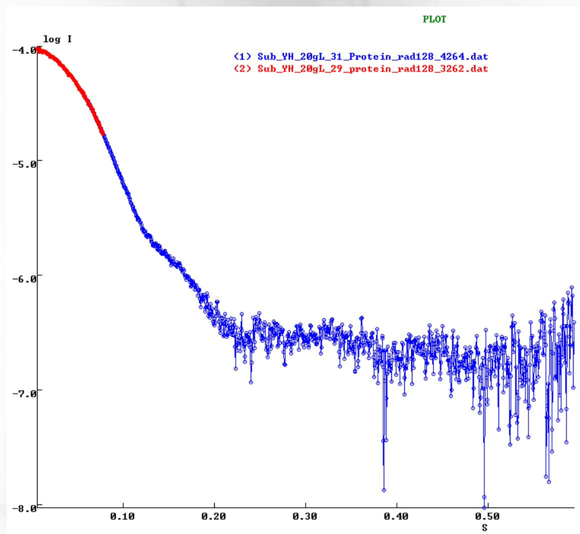
AutoSampler

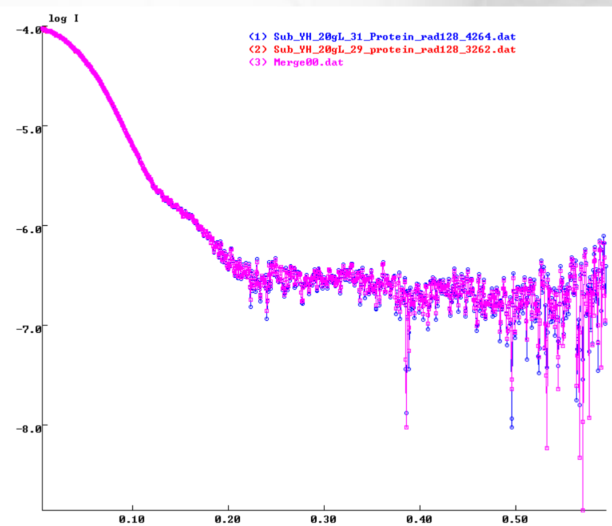






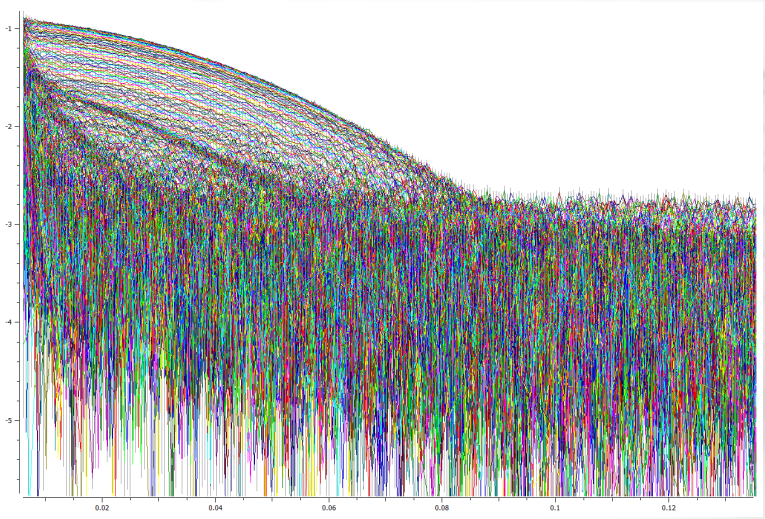


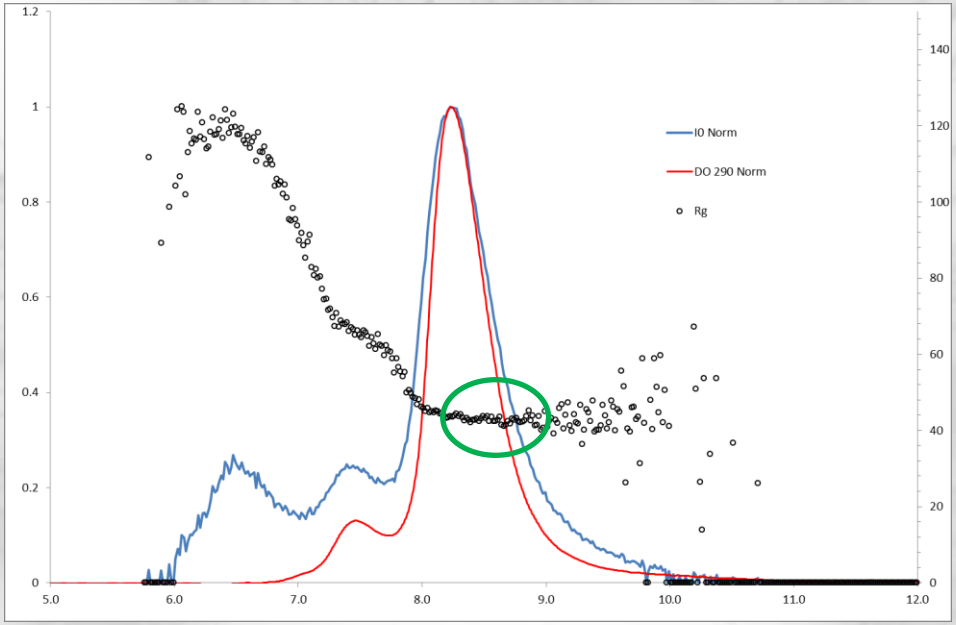


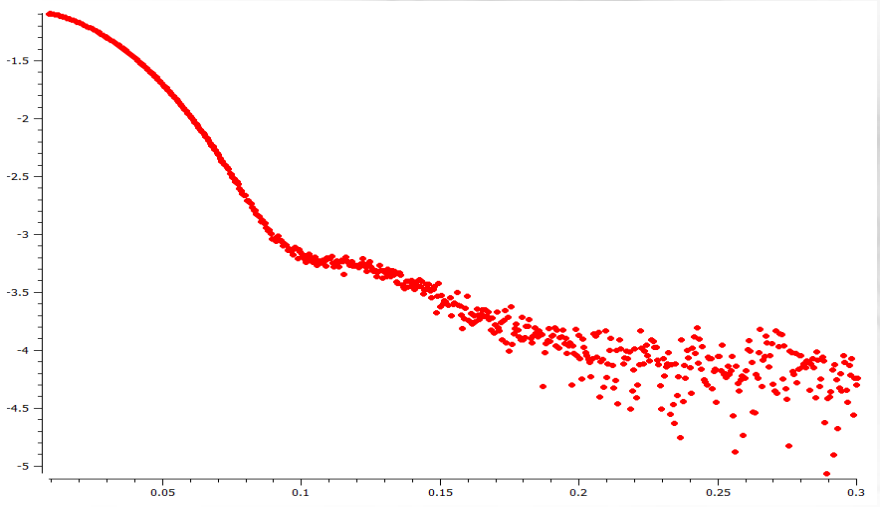


SEC-SAXS

SEC-SAXS

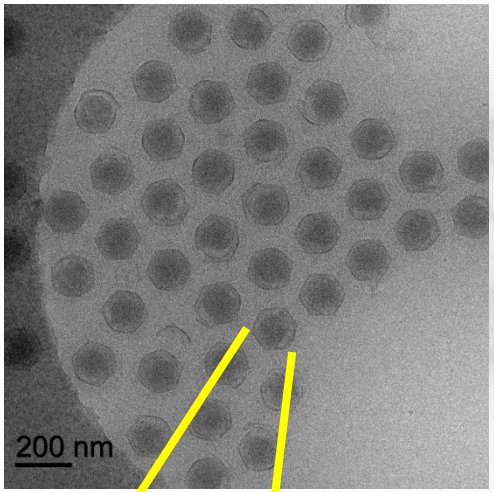


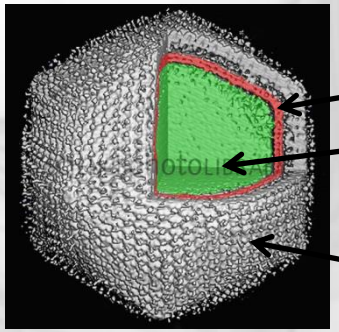




THE BEGINNING OF A NICE STORY

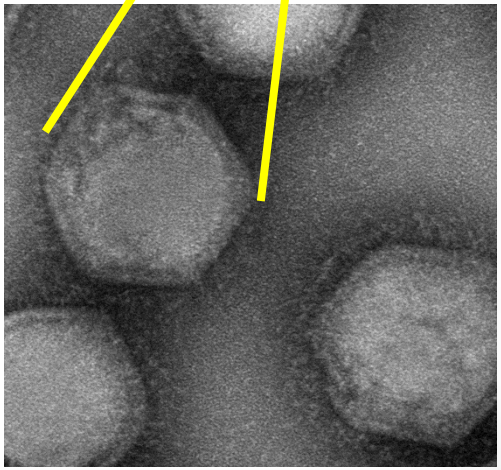
Chilo Iridescent Virus (CIV)





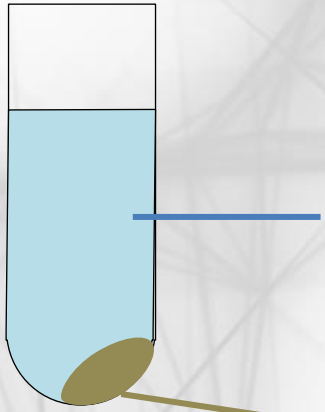
Vesicule:
- Lipid bilayer
- DNA + Protein

Capside:
Major Capsid Protein (MCP)
Protéines fibrillaires
Minor capsid proteins



UltraCentrifugation

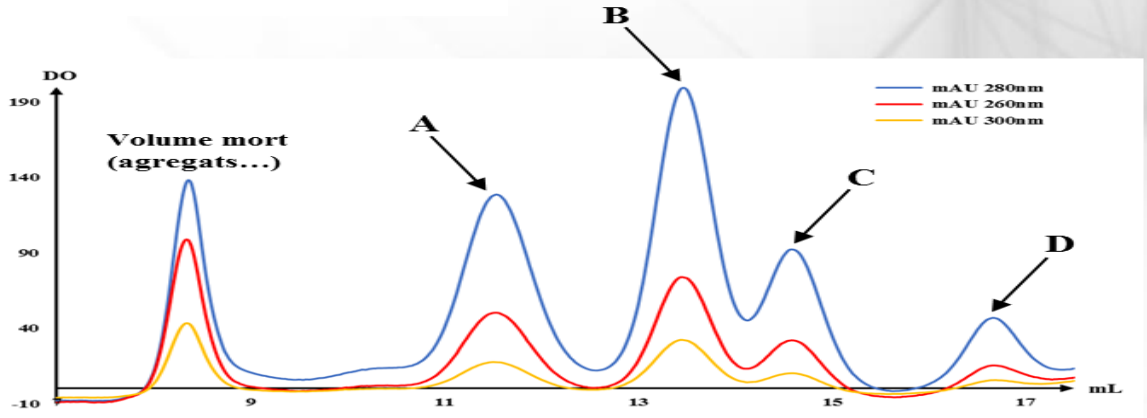
Capside destruction



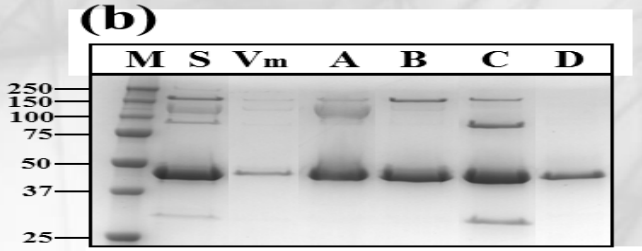
Pierre Montaville – Synchrotron Soleil

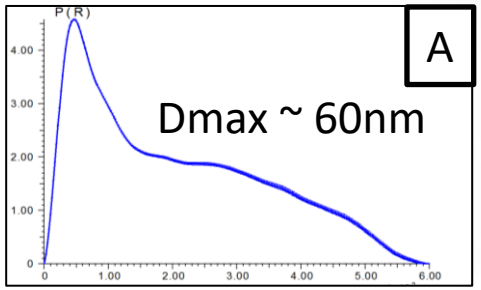
Chilo Iridescent Virus (CIV)

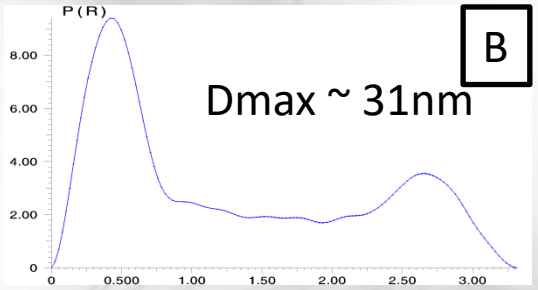
Gel filtration

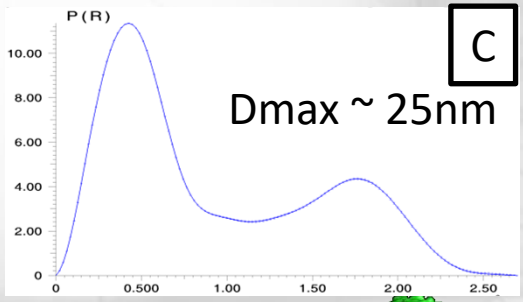


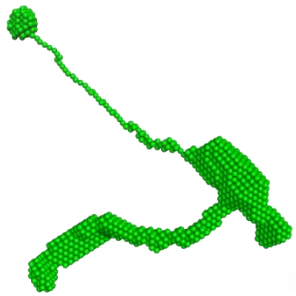
SDS + mass spectrometry

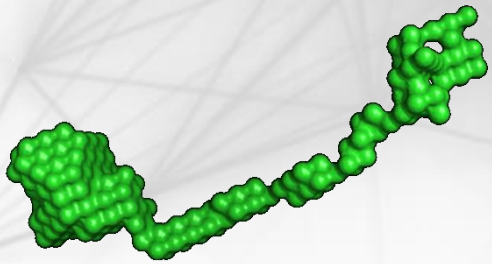


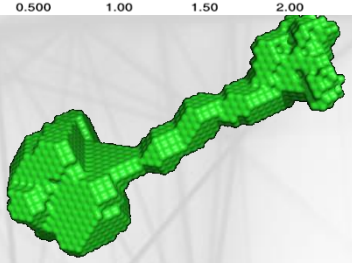




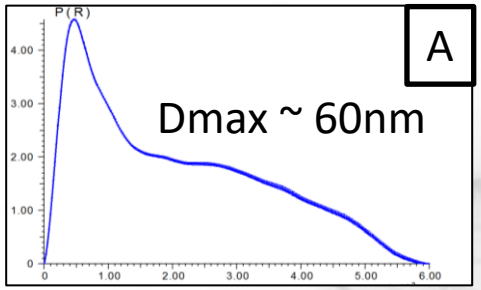


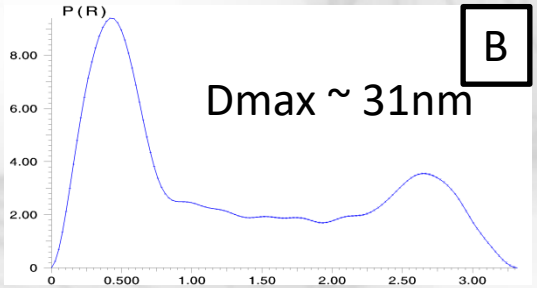


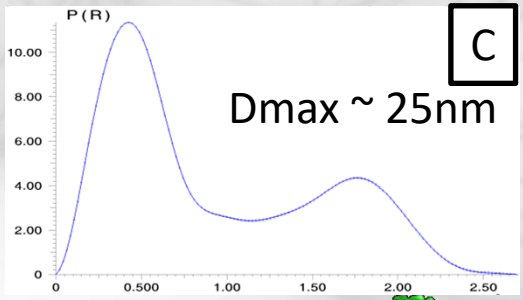


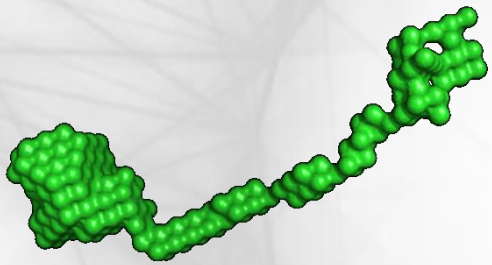


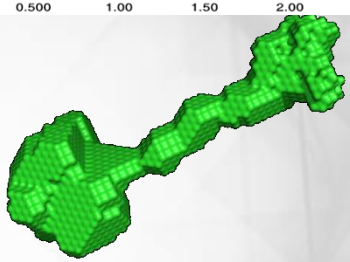
Chilo Iridescent Virus (CIV)

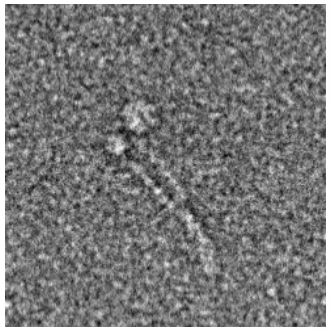


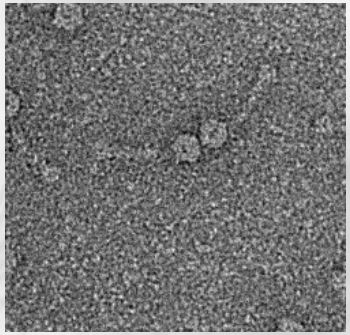


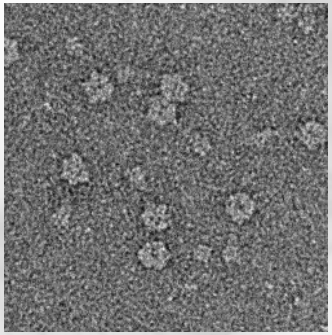


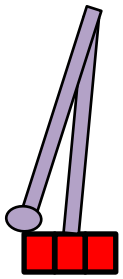


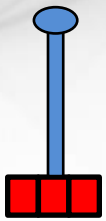


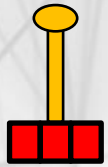












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