Project CAPSID
of the Slovakia-Austria Cooperation Programme

EUROPEAN UNION

Interreg
Slovakia-Austria
European Regional Development Fund

PREDNÁŠKA

'Seminar Speaker Series'

s občerstvením

prednáša

Dr. Bertrand RAYNAL

Research Engineer for Molecular Biophysics at Institute Pasteur, Paris

Structural biology is moving forward …

HydEnGe: A Program for *ab initio* shape determination using Hydrodynamic Parameters acquired on Benchtop Biophysical Instruments

6.11.2019 o 13:00

veľká zasadačka Virologický ústav BMC SAV

Organized by:

BIOMEDICÍNSKE CENTRUM SLOVENSKÁ AKADÉMIA VIED

Vienna BioCenter
CORE FACILITIES

Project CAPSID
of the Slovakia-Austria Cooperation Programme

EUROPEAN UNION

Interreg
Slovakia-Austria
European Regional Development Fund

Seminar Speaker

Dr. Bertrand RAYNAL

Research Engineer for Molecular Biophysics at Institute Pasteur, Paris

Institut Pasteur

Bertrand Raynal
Bâtiment 67
25 rue du Dr Roux
75724 Paris cedex 15

+ 33 (0)1 44 38 94 75
bertrand.raynal@pasteur.fr

RESEARCH PROJECTS

- **Biophysical characterization of the translocation process** of Bordetella pertussis adenylate cyclase toxin CyaA - This project aims to describe the hydrodynamic and thermodynamic properties of the CyaA toxin and the conformational changes it undergoes during its translocation process across the lipid membranes of Bordetella pertussis (secretion) and of its eukaryotic target cell (intoxication).
- **Molecular crowding** - We are interested in investigating the effects of molecular crowding, i.e., excluded volume effects, mimicking the effects of high concentrations of macromolecular solutes found in biological fluids on biochemical properties of proteins.
- **In solution conformation of the SigmaS subunit of RNA polymerase** - There is no tridimentional structure available for free sigma factors. We have recently probed the conformation of SigmaS in solution by analytical ultracentrifugation, small angle X-ray scattering and Hydrogen-Deuterium exchange coupled with mass spectrometry.
- **CyaA secretion, folding and translocation across membrane** - One challenging aspect of the structural and biophysical studies of CyaA arises from the complexity of this toxin, a large (1706 amino-acids) multi-domain protein that is post-translationally acylated and exhibits a pronounced hydrophobic character limiting its solubility.

BIO

1999 - MSc in biochemistry, Université Paul Sabatier Toulouse III ERASMUS training period at the University of Manchester; **2003 - PhD** in biochemistry University of Manchester
Since 2007 - Engineer in charge of the “hydrodynamic characterization of macromolecules” section, Centre of Biophysics of Macromolecules and their Interactions (PFBMI), Institut Pasteur, Paris; **2004-2007** - Postdoctoral Research associate University of (supervisors: Prof. Cay Kielty and Dr Adrian Shuttleworth) Manchester, UK Fibrillin structure and its organisation in microfibrils; **2003-2004** - Postdoctoral Research associate (supervisor: Dr Dave Thornton) University of Organisation of normal and pathological mucus (cystic fibrosis, asthma…) and Manchester, UK influence of calcium; **1999-2003** - Research Assistant (supervisors: Prof. John Sheehan and Prof. Tim Hardingham) University of The molecular structure, organisation and interactions in mucus gels. Manchester, UK

Organized by:

BIOMEDICÍNSKE CENTRUM SLOVENSKÁ AKADÉMIA VIED

Vienna BioCenter
CORE FACILITIES

SEMINAR ANNOTATION

HYDENGE: A PROGRAM FOR AB INITIO SHAPE DETERMINATION USING HYDRODYNAMIC PARAMETERS ACQUIRED ON BENCHTOP BIOPHYSICAL INSTRUMENTS

Bertrand Raynal

Structural biology is moving forward in an accelerated way with the rise of electron microscopy, the automation of synchrotron beamlines, and new HPLC-SAXS setups. However, all these approaches encounter difficulties when faced with complex samples such as multiprotein assemblies and membrane proteins…It is therefore essential to validate their proposed spatial arrangement with other techniques and to propose simpler approaches to determine the shape of these structures. We have developed a new program (HydEnGe) that generates shapes using a combination of hydrodynamic measurements that can be performed using bench top biophysical instruments, and do not require access to very large instrumentation. The program uses data input such as Sedimentation coefficient, Intrinsic viscosity, Hydrodynamic radius, and Radius of gyration that can be measured in most molecular biophysics laboratories. All these parameters are characteristic of the spatial arrangement of the molecules, and are influenced in different ways by features of the molecules of interest such as the solvent-accessible surface, the molecular mass and its distribution, the size … Consequently, the number of solutions for the spatial arrangement of the molecule is limited, and give solution that has overall similar shape. Using a simulated annealing algorithm, molecules are represented as an assembly of spheres, from which hydrodynamic parameters are calculated using the Hydropro software and compared to experimental values. The sphere assembly is then randomly modified and the calculation process is re-iterated. If the modification allows a better fitting to the experimental values the model is then used as the starting point for a new modification. Step by step the model converges towards a solution in which the calculated hydrodynamic parameters are in close agreement with the experimental value. After using HydEnGe several times with the same hydrodynamic experimental values, an average ab initio shape is generated. The method will be illustrated with an ab initio modeling of lysozyme and of others proteins, compared to known structures (see figure). The limit of the ab initio shape will be presented using modelisation of define structure… The use of HydEnGe to estimate the shape of membrane proteins and large assemblies will be discussed, as well as its application to improve the results obtained by structural biology techniques. Overall the new HydEnGe program could be implemented in a close future in many institutions possessing molecular biophysic instruments and wishing to integrate the experimental data obtained to improve their knowledge about the shape and conformation of the molecules there are characterizing.

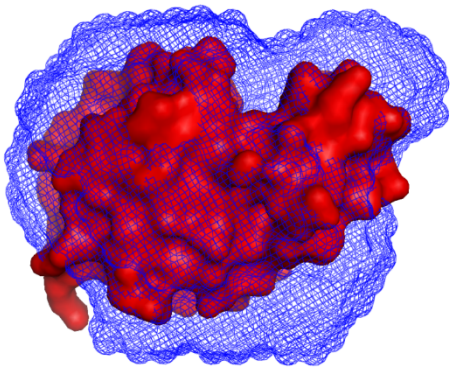

Figure: Hydenge envelope of lyzoyme (blue) compare to unhydrated structure (red)

Organized by: