NMR Meets Biology

Kerala 2016

Book of Abstracts

ssNMR in Comparison to Other Structural Investigation Techniques

Beat Meier
Department of Chemistry and Applied Biosciences, ETH Zürich, Switzerland

Abstract

The lecture will show how solid-state NMR can be combined with other structural methods, notably electron microscopy and x-ray crystallography. After an introduction to the topic, we will show this at the example of the ASC-PYD filaments, as well as the DnaB Helicase and its interaction partners.

Date: 14 January 2016 **Time: 16:00-17:00**

Future Challenges in Structural Biology

Hartmut Oschkinat
Leibniz-Institut für Moleculare Pharmakologie, Berlin, Germany

Abstract

Date: 14 January 2016 **Time: 17:30-18:30**

Practical Aspects of Solid State NMR

Gerhard Althoff
Bruker BioSpin GmbH, Silberstreifen, Germany

Abstract

This contribution focusses on the hardware needed for of solid-state NMR. As the vast majority of solid-state NMR experiments is performed under magic angle sample spinning (MAS) the emphasis is on MAS probes with respect to:

- MAS rotor and stator design
- Properties of air bearings
- Ancillary effects of sample spinning: frictional heating and centrifugal forces
- Rotor handling and precautions
- Probe RF design for double and triple resonance experiments
- Probe channel insulation and external filters
- Ancillary effects of RF pulses: RF sample heating and coil designs to minimize this effect
- Special probe designs for low or high temperatures and for dynamic nuclear polarization (DNP)

Date: 14 January 2016 **Time: 18:30-19:30**

The Hamiltonian Operator

Beat Meier
Department of Chemistry and Applied Biosciences, ETH Zürich, Switzerland

Abstract

The basics of the quantum description of NMR spectra using spherical tensor operators will be introduced and applied to static and MAS experiments in two and three dimensions.

Date: 15 January 2016 **Time: 08:00-09:20**

Exercise Hour

Beat Meier
Department of Chemistry and Applied Biosciences, ETH Zürich, Switzerland

Abstract

This will an exercise hour based on the above session.

Date: 15 January 2016 **Time: 09:40-11:10**

Time-dependent Hamiltonians: AHT and Floquet Theory

Mathais Ernst
Department of Chemistry and Applied Biosciences, ETH Zürich, Switzerland

Abstract

Generating different effective Hamiltonians during various time periods of experiments is one of the key reasons for the large number of experiments that can be realized in solid-state magic-angle spinning NMR. Although the laboratory-frame spin-system Hamiltonian is time independent, magic-angle spinning as well as radio-frequency irradiation lead to a time dependence of the Hamiltonian. Therefore, methods to treat time-dependent Hamiltonians are essential for understanding solid-state MAS NMR experiments. In this lecture the origin of time-dependent Hamiltonians as well as the two main ways to treat them, namely average Hamiltonian theory and Floquet theory, are discussed. Simple examples will be used to illustrate and compare the two methods.

Date: 15 January 2016 **Time: 11:30-13:00**

Spin Dynamics

Malcolm H. Levitt

University of Southamptom, United Kingdom

Abstract

In most forms of NMR we deal with nuclear spin systems which are very close to thermal equilibrium. However there are several phenomena which involve highly non-equilibrium nuclear spin systems, and which are of great interest since they may be associated with strongly enhanced NMR signals, sometimes by many orders of magnitude. These are: (1) hyperpolarization effects, in which samples are prepared with nuclear spin temperatures which may be much smaller than that of the environment; (2) spin isomerism, which entangles the spatial and spin quantum states, investing some nuclear spin states with energies which greatly differ from ordinary nuclear Zeeman energies, and which allows the ready preparation of samples in highly non-equilibrium spin states. I expect to improvise a discussion of the various phenomena involving nuclear spins far from equilibrium, the relationship between them, and the field of long-lived nuclear spin states.

Date: 15 January 2016 **Time: 14:30-16:30**

Dynamics of Membrane Proteins

Daniel Huster

Institut für Medizinische Physik und Biophysik, Universität Leipzig, Germany

Abstract

Membrane proteins are highly dynamic molecules that undergo numerous dynamic conformational transitions with varying amplitudes within a broad window of correlation times. In the presentation, I will provide a summary on the dynamic modes of membrane proteins and an overview of solid-state NMR methods that allow to study these motions. From the general classification of motions observed on the NMR time scale (fast ($\tau<\mu$s), intermediate ($\tau \sim \mu$s), slow motions ($\tau>\mu$s)) some well-established methods for the elucidation of membrane protein dynamics will be discussed. These include the study of motionally averaged anisotropic interactions detected in separated local field experiments, relaxation time measurements and analysis, line shape analysis for intermediate time scale motional analysis, and exchange spectroscopy to study slow (millisecond timescale) protein.

Date: 15 January 2016 **Time: 17:00-18:00**

RNA recognition motifs: A story of one fold with different partners

Neel Sarovar Bhavesh

International Centre for Genetic Engineering and Biotechnology, New Delhi, India

Abstract

RNA binding proteins or RBPs play an indispensable role in cellular machinery, especially processes such as transcription, post-transcriptional modification of RNA, RNA transport and stabilization, among others. Post-transcriptional modifications of RNA are a major route through which eukaryotes regulate gene expression. These modifications include splicing, mRNA polyadenylation, 5 capping and RNA editing. RBPs bind RNA through specific RNA binding domains (RBDs) or modules. Binding affinities and specificities vary throughout this family of proteins. The RNA recognition motif or RRM is the most widely distributed RBD in nature. RRMs are canonically identified through the presence of two RNA binding consensus motifs (RNP). They have a common three-dimensional architecture, which classically consists of a four-stranded β-sheet supported by two α-helices, with the β-sheet serving as the major surface for RNA recognition.

In spite of these unifying traits of RRMs, they possess a remarkably diverse RNA recognition capability. Our recent delineation of the RNA-binding mechanism of human TAF15 protein highlights how the concave face of its carboxy terminal RRM recognizes structured loop elements on RNA, in a non-canonical manner. Mutations in this protein have been implicated in familial amyotrophic lateral sclerosis or FALS. The human ETR3 protein on the other hand, possesses three RRMs, which sequence-specifically recognizes CUG and UG-rich RNA. This is carried out through specific interaction of a uracil base by a cleft on the β-sheet surface. The N-terminal RRM of malarial SR1 alternative splicing factor that semi-specifically recognizes pyrimidines exhibits a similar mode of interaction. We have delineated how the latter two instances represent a canonical form of RRM-RNA interaction, with $\pi - \pi$ interactions between aromatic amino acids and nucleotide bases being responsible for binding. On the other hand, the TAF15-RRM-RNA interface is largely dominated by hydrogen bonding between charged amino acids and polar groups on the RNA. Our work on RNA recognition by RRMs thus paints an interesting picture of how a single fold is able to recognize different cognate RNAs by virtue of minor but crucial alterations to its binding surface. In addition, delineation of RNA binding specificities of RRMs has provided molecular clues to the progression of debilitating diseases such as myotonic dystrophy, FALS and malaria.

Date: 15 January 2016 **Time: 18:00-18:30**

Dock-lock mechanism of β-amyloid fibril formation in membrane

Anirban Bhunia

Bose Institute, Kolkata, India

Abstract

Understanding the mechanisms of biological processes requires precise knowledge of the three-dimensional structures of the executor molecules such as proteins, bioactive peptides and others. Atomic-resolution structures of well-folded proteins or complexes can be obtained from X-ray crystallography. However, a large number of proteins or domains of large proteins (e.g., in signaling cascades) and bioactive peptides (e.g., antimicrobial peptides, amyloid peptide) appear to be dynamic, thus limiting the application of X-ray-based methods. On the other hand, gaining insights into such molecular systems at the atomic level is possible using nuclear magnetic resonance (NMR) spectroscopy. The functional aspect of neurodegenerative diseases linked with amyloid beta (Aβ_{40} / Aβ_{42}) fibril formation is well known from the in vitro studies. However, membrane could play an important role for this fibril formation. To date, there is a paucity of information detailing the interaction of amyloid beta (Aβ_{40} / Aβ_{42}) proteins with membrane structures. In this talk I shall focus on elucidating the biophysical properties of the interactions between amyloid beta (Aβ_{40} / Aβ_{42}) proteins and model membrane system.

Date: 15 January 2016 **Time: 18:30-19:00**

Dipolar Decoupling

P. K. Madhu
Tata Institute of Fundamental Research, Mumbai, India

Abstract

Dipolar decoupling is essential in proton-rich systems to get high-resolution solid-state NMR spectra of either the rare spins, such as 13C, or protons. The former requires heteronuclear decoupling pulse methods and the latter needs homonuclear decoupling pulse schemes. This talk will address both the issues and introduce state-of-the-art methods in both the areas.

Date: 16 January 2016 **Time: 08:00-09:20**

MAS NMR Methods for Structural and Dynamics Analysis of Biological Assemblies: Sensitivity-Enhanced Rapid Data Collection through Nonuniform Sampling and Site-Specific Measurements of Dipolar/CSA Tensors

Tatyana Polenova
University of Delaware, Newark, USA

Abstract

MAS NMR spectroscopy is an essential method for structural and dynamics analysis of biological systems and is particularly powerful for atomic-resolution studies of large biomolecular assemblies where other structural techniques provide limited information. In MAS NMR studies of biomolecular assemblies, sensitivity and resolution remain a main challenge. I will discuss the principles and applications of nonuniform sampling (NUS) as one approach for gaining bona fide time-domain sensitivity enhancements. I will present an overview of theory and experiments demonstrating that with the use of appropriately designed NUS sampling schedules, sensitivity gains of 1.7 - 2 fold are attained without the compromise in resolution. These sensitivity enhancements are compounded in each indirect dimension, resulting in considerable time savings and enabling collection of nD MAS NMR spectra in systems where traditional linear sampling data collection is prohibitively time consuming. I will discuss the maximum entropy interpolation method (MINT) introduced by us for processing of NUS data that permits quantification of sensitivity. I will demonstrate that NUS is compatible with paramagnetic-relaxation-assisted condensed data collection (PACC) under fast MAS conditions, a combined NUS-PACC approach, yielding dramatic, 20-26 fold time enhancements in heteronuclear-detected 3D experiments. In the second part of the lecture, I will discuss approaches for site-specific determination of heteronuclear dipolar, heteronuclear CSA, and ^{1}H CSA interactions. RN-symmetry based sequences originally introduced by Malcolm Levitt and colleagues, are a powerful framework in the design of experiments for recoupling desired anisotropic tensorial interactions. These experiments are ideally suited for site-specific measurements of dipolar and CSA tensors in biological systems, including biological assemblies, and are compatible with a broad range of MAS conditions, including spinning frequencies of 40 kHz and above. I will discuss the application of this approach to the determination of molecular dynamics in HIV-1 capsid assemblies through the measurements of backbone dipolar interactions, where, in conjunction with MD simulations, we have obtained a comprehensive view of the molecular motions. I will also discuss the measurements of ^{1}H CSA tensors in a microtubule-associated CAP-Gly protein, and the connection of the determined ^{1}H CSA parameters to the hydrogen bonding interactions.

Date: 16 January 2016 **Time: 09:40-11:10**

Assignments/Structural Restraints from Multidimensional Spectra

Anja Böckmann
Institut de Biologie et Chimie des Protéines, Lyon, France

Abstract

Protein building principles will be introduced. Sequential assignments based on 3D experiments will be illustrated, and the measurement of structural restraints using 13C and 15N correlation spectra will be introduced.

Date: 17 January 2016 **Time: 08:00-09:20**

Calculation of NMR parameters using Gaussian

Tatyana Polenova
University of Delaware, Newark, USA

Session Requirements
Gaussian v9.x Viewer installed on your laptop.
Date: 17 January 2016 **Time: 09:40-10:40**

Calculation of NMR parameters using Gaussian: Tutorial Session

Tatyana Polenova
University of Delaware, Newark, USA

Session Requirements
Gaussian v9.x Viewer installed on your laptop.
Access to Gaussian09 will be provided via SSH.
Date: 17 January 2016 **Time: 11:30-13:00**

Workshop 1 - Parallel Session 1 Hamiltonians and Rotations

Mathais Ernst
Department of Chemistry and Applied Biosciences, ETH Zürich, Switzerland

Date: 18 January 2016 **Time: 14:30-16:30**

Workshop 1 - Parallel Session 2 NMR Assignments

Anja Böckmann
Institut de Biologie et Chimie des Protéines, Lyon, France

Session Requirements
CCPNMR installed on your laptop. Example datasets will be provided.
Date: 18 January 2016 **Time: 14:30-16:30**

Observation of long-range H_N-H_N/Hα-Hα contacts by frequency-selective recoupling in fully protonated proteins at 100 kHz MAS

Vipin Agarwal
TIFR Centre for Interdisciplinary Sciences, Hyderabad India

Abstract

Deuterated proteins with different degree of protonation have been used successfully used in tertiary structure determination of proteins by solid-state NMR. However, not all proteins can be deuterated. So, new methods are required to structurally characterize non-deuterated biomolecules. The progress in magic-angle spinning (MAS) technology now provides a possibility to record meaningfully resolved proton spectrum of fully protonated proteins. Fast MAS (100 kHz) combined with high static fields has enabled protondetected correlation spectroscopy in fully protonated samples with sufficient resolution, primarily for the purpose of assignment. Here, we propose a new experimental method to obtain long-range proton-proton contacts in fully protonated proteins at fast MAS. The novel method demonstrates that ^{1}H-^{1}H contacts on the order of 6-7 Åcan be obtained in fully protonated proteins. A systematic comparison of the experimental 1H-1H contacts was performed with the expected 1H-1H contacts at a distance of 7 Åbased on the X-ray structure

Date: 17 January 2016 **Time: 17:00-17:30**

Understanding the role of intrinsic dynamics in substrate recognition by Skp1 and the F-box domain in the SCF complex

Ashutosh Kumar
Indian Institute of Technology Bombay, Mumbai, India

Abstract

Precisely regulated protein function is vital for most of the cellular processes. Such fidelity in regulation is required for prevention of premature and adverse interactions among various signaling cascades in cellular compartments to ensure that binding is spatially and temporally appropriate. NMR spectroscopy along with molecular dynamics simulations appear as elegant tools for characterization of biological macromolecules with its unique ability to provide a coherent insight into the structure and intrinsic dynamics. Here, we have investigated the dynamic regulation of proteins involved Ubiquitin-dependent proteolysis (UPS) machinery. UPS regulates protein abundance, and in-turn serves as a central regulatory function in many biological processes. The SCF (Skp1CullinF-box protein) complex is one such macromolecular assembly that ubiquitinates a broad range of proteins involved in cell cycle progression, signal transduction and transcription. In SCF complex, Skp1 is a crucial adaptor protein which directly interacts with F-box proteins and cullin-1.This complete assembly is responsible for targeting proteins for the ubiquitin mediated degradation. Using NMR spectroscopy, we investigated dynamics regulation by adapter protein Skp1, and with MD simulation we probed substrate regulation in complete SCF assembly. We show that the conformational behaviour of the F-box domain of Skp2 and the domain motions of Skp1 and Skp2 proteins in the binary (without substrate) and ternary (with substrate) complexes is modified and adapted depending upon the target substrate to facilitate the ubiquitination of the substrate molecule.

Date: 17 January 2016 **Time: 17:30-18:00**

Behavior of Water in Cortical Bones

Neeraj Sinha
Centre for Biomedical Research, Lucknow, India

Abstract

The mechanical properties of cortical bone, which is largely composed of collagen, hydroxyapatite, and water, are known to hinge on hydration. Recently, the characteristics of water in bone have drawn attention as potential markers of bone quality. We will present the dynamics, diffusion, population, and exchange of water in cortical bone by NMR relaxation and diffusion methodologies. Relaxation measurements over time scales ranging from 0.001 to 4.2 s reveal two distinguishable water environments. Systematic exposure to ethylenediaminetetraacetic acid or collagenase reveals one peak in our 2D relaxation map belonging to water present in the hydroxyapatite-rich environment and a second peak with shorter relaxation times arising from a collagen-rich site. Diffusion-T2 measurements allowed for direct measurement of the diffusion coefficient of water in all observable reservoirs. Further, deuterium relaxation methods were applied to study cortical bone under an applied force, following mechanical wear or fracture. The tumbling correlation times of water reduce in all three cases, indicating that water dynamics may be used as a probe of bone quality. Lastly, changes in the relative populations and correlation times of water in bone under an applied force suggest that load bearing occurs largely in the collagen- rich environment and is reversible.

Date: 17 January 2016 **Time: 18:00-18:30**

^{13}C NMR Investigations of Neurotransmitter Energetics in Alzheimer's Disease

Anant Patel
University of Delaware, Newark, USA

Abstract

Alzheimers disease (AD) is one of the most common neurodegenerative disorders, characterized by progressive memory impairment and loss in cognitive functions. Although, AD was discovered more than a century ago, there is neither a quantitative diagnosis nor cure of the disease exist. Glutamate and α-aminobutyric acid (GABA) are the major excitatory and inhibitory neurotransmitters, respectively, in the matured nervous system.

These neurotransmitters play major roles in energy metabolism, cortical excitability and cognitive function. Dysfunctions in glutamate and GABA neurotransmitters pathways are associated with many neurological and neuropsychiatric disorders. 13C NMR studies in rats brain have established that neurotransmitter energetics is supported by oxidative glucose metabolism[1,2]. We are using 1H-[13C]-NMR spectroscopy in conjunction with infusion of 13C labeled substrates (glucose and acetate) to evaluate the neurotransmitters energetics with the progress of age in mouse model of AD[3]. A three-compartment metabolic model is used to estimate different metabolic rates from the 13C turnover of amino acids (GluC4/C3, GABAC2/C3, AspC2/C3 and GlnC4/C3) from [1,6-13C2]glucose[4]. Our analysis indicated that cortical and hippocampal regions, responsible for cognitive and memory functions, exhibit profound reduction in glutamatergic and GABAergic metabolic activity at high plaque loading in APP-PS1 mice. In contrast, astroglial activity was found to be enhanced in AD mice. In addition, APP-PS1 mice exhibit severe decline in the flux through pyruvate carboxylase and pentose phosphate pathways[5]. The reduction in these fluxes would lead to a compromised defense system for ammonia detoxification and antioxidant capacity in AD. Most interestingly, we found that neuronal metabolism was impaired even at the preclinical stage suggesting potential of 13C NMR based metabolic measurements for the early detection of AD. In this presentation, I will be discussing these findings in detail.

Date: 17 January 2016 **Time: 18:30-19:00**

Paramagnetic Shifts and Paramagnetic Relaxation Enhancements

Guido Pintacuda
Institut des Sciences Analytiques, Lyon, France

Abstract

Paramagnetic centers originate from unpaired electrons that are intrinsic features of organic radicals and of many transition metal ions. In this lecture, we will illustrate how unpaired electrons affect the NMR spectrum of the surrounding nuclear spins, namely by altering their chemical shifts and by increasing their relaxation rates. The effects arising from paramagnetism depend in a well-defined manner on th e electronic configuration of the metal and on the structure and dynamics of the molecule, providing information on the electronic states of the paramagnetic center, as well as a number of structural restrai nts with wide ranging implications for molecular and biological sciences.

Date: 18 January 2016 **Time: 08:00-9:20**

Hyperpolarization

Mathais Ernst
Department of Chemistry and Applied Biosciences, ETH Zürich, Switzerland

Date: 18 January 2016 **Time: 09:40-10:40**

CYANA: A Tool for Structure Calculations

Peter Güntert
Goethe University Frankfurt am Main, Frankfurt am Main, Germany

Abstract

NMR structure determination relies on distance restraints and other conformational restraints obtained from assigned solution- or solid-state NMR spectra. The CYANA program package is a widely used tool for calculating the three-dimensional structures of proteins and nucleic acids on the basis of NMR restraints. Simulated annealing driven by molecular dynamics simulation in torsion angle space is the fundamental algorithm that enables efficient NMR structure calculations in CYANA. The most important input are distance restraints from NOESY or corresponding solid-state NMR spectra, H-bonds, paramagnetic relaxation enhancement or EPR measurements. Additional types of conformational restraints can be used: torsion angle restraints, residual dipolar couplings (RDC), pseudo-contact shifts, and symmetry restraints.

The CYANA program package also provides an algorithm for the automated assignment of cross peaks in NOESY or corresponding solid-state NMR spectra that can in most cases replace the very cumbersome manual assignment of these cross peaks, thereby saving weeks or months of the spectroscopists time.

Also the resonance assignment, which has to precede the collection of distance restraints, constitutes, a time-consuming step towards a structure. To automate this resonance assignment step, CYANA includes the FLYA algorithm for the automated assignment of backbone and side-chain chemical shifts. FLYA has the reliability and flexibility to replace manual assignment procedures for most NMR studies of proteins4, including those analyzed by solid-state NMR.

CYANA has recently been used for determining atomic resolution structures of several amyloid fibrils by solid-state NMR, e.g. those of the Aβ(1-40) peptide involved in Alzheimers disease. This application will also be presented in the practical session.

A long-standing question in protein NMR is the lack of a reliable measure of structural accuracy, i.e. how close NMR conformers are to the true structure. Instead, the precision of structure bundles is widely (mis)interpreted as a measure of structural quality. Attempts to increase precision often overestimate accuracy by tight bundles of high precision but much lower accuracy. To overcome this problem, a new protocol for NMR structure determination with the software package CYANA will be presented that produces, like the traditional method, bundles of conformers in agreement with a common set of conformational restraints, however with a realistic precision that is, throughout a variety of proteins and NMR data sets, a much better estimate of structural accuracy than the precision of conventional structure bundles.

Date: 18 January 2016 **Time: 11:00-13:00**

Workshop 2 - Parallel Session 1
Structure Calculation

Peter Güntert
Goethe University Frankfurt am Main, Frankfurt am Main, Germany

Abstract

This session will be based on teh previous talk and will include the structure calculation of the Aβ(1-40) peptide.

Session Requirements
CYANA installed on your laptop

Date: 18 January 2016 **Time: 14:30-16:30**

Workshop 2 - Parallel Session 2
SIMPSON

There will be two parallel tutorials on SIMPSON:

- Introductory tutorial (P.K.Madhu). This tutorial will cover the basics of setting up simulations in SIMPSON.
- Advanced tutorial (Asif Equbal and Ravi Shankar). This session will cover advanced topics such as setting up of 2D optimizations, usage of shape-files, curve-fitting and optimal control theory.

Session Requirements
SIMPSON installed on your laptop

Date: 18 January 2016 **Time: 14:30-16:30**

Understanding the Conformational Dynamics of the T4 Lysozyme L99A Cavity Mutant

Pramodh Vallurupalli
TIFR Centre for Interdisciplinary Sciences, Hyderabad, India

Abstract

In solution T4 Lysozyme L99A (T4L L99A) interconverts between two conformers with the minor conformer having a population of 3% and a lifetime of 1 millisecond at 20 oC. The structure of this minor conformer has been obtained using CPMG NMR experiments. The major and minor state structures are very similar to one another, with only Phe114 that is solvent exposed in the major form getting buried into the cavity in the minor form. While we have structures of the two endpoints of the transition, there are still several open questions regarding the dynamics itself. Where does the protein get the energy to surmount the activation barrier? What is the size of the activation barrier that separates these two states? In what regime does the dynamics lie, Kramers or Hinshelwood? Results from our recent NMR and computational studies that address these questions will be presented.

Date: 18 January 2016 **Time: 17:00-17:30**

Steric crowding of the turn region alters the tertiary fold of Amyloid-β_{18-35} and makes it soluble

Venus Singh Mithu
Guru Nanak Dev university, Amritsar, India

Abstract

Aβ self-assembles into parallel cross-β fibrillar aggregates, which is associated with Alzheimers disease pathology. A central hairpin turn around residues 23-29 is a defining characteristic of Aβ in its aggregated state. Major biophysical properties of Aβ, including this turn, remain unaltered in the central fragment Aβ18-35. Here, we synthesize a single deletion mutant G25 with the aim of sterically hindering the hairpin turn in Aβ18-35. We find that the solubility of the peptide goes up by more than twenty fold. Though some oligomeric structures do form, solution-state NMR spectroscopy shows that they have mostly random coil conformations. Fibrils ultimately form at a much higher concentration, but have widths about twice that of Aβ18-35, suggesting an opening of the hairpin bend. Surprisingly, 2D solid-state NMR shows that the contact between F19 and L34 residues, observed in full length Aβ and Aβ18-35, is still intact in these fibrils. This is possible if the monomers in the fibril are arranged in an antiparallel β-sheet conformation. Indeed, IR measurements, supported by tyrosine cross-linking experiments, provide a characteristic signature of the antiparallel β-sheet. We conclude that the self-assembly of Aβ is critically dependent on the hairpin turn, and on the contact between the F19 and L34 regions, making them potentially sensitive targets for Alzheimers therapeutics. Our results show the importance of specific conformations in an aggregation process thought to be primarily driven by non-specific hydrophobic interactions.

Date: 18 January 2016 **Time: 17:30-18:00**

Multiple sequential acquisitions using one and two receivers to speed up data acquisition in MAS solid state NMR

Kaustubh R. Mote
TIFR Centre for Interdisciplinary Sciences, Hyderabad, India

Abstract

One of the fundamental challenges in the application of solid state NMR is its limited sensitivity, yet a majority of experiments do not make efficient use of the limited polarization available. The loss in polarization in a single acquisition experiment is mandated by the need to select out a single coherence pathway. In contrast, sequential acquisition strategies can encode more than one pathway in the same experiment or recover unused polarization to supplement a standard experiment. I will present pulse sequences that implement sequential acquisition strategies on one and two receivers with a combination of proton and carbon detection to record multiple experiments under magic angle spinning. The pulse sequence suite is further expanded by including pathways that allow the recovery of residual polarization, the so-called "afterglow" pathways, to efficiently encode a number of pulse sequences to aid in assignments and chemical shift mapping.

Date: 18 January 2016 **Time: 18:00-18:30**

Investigating the early-misfolding events in transthyretin amyloidosis

Sujoy Mukherjee
Structural Biology and Bioinformatics Division,
CSIR - Indian Institute of Chemical Biology, Kolkata, India

Abstract

One of the fundamental challenges in the application of solid state NMR is its limited sensitivity, yet a majority of experiments do not make efficient use of the limited polarization available. The loss in polarization in a single acquisition experiment is mandated by the need to select out a single coherence pathway. In contrast, sequential acquisition strategies can encode more than one pathway in the same experiment or recover unused polarization to supplement a standard experiment. I will present pulse sequences that implement sequential acquisition strategies on one and two receivers with a combination of proton and carbon detection to record multiple experiments under magic angle spinning. The pulse sequence suite is further expanded by including pathways that allow the recovery of residual polarization, the so-called "afterglow" pathways, to efficiently encode a number of pulse sequences to aid in assignments and chemical shift mapping.

Date: 18 January 2016 **Time: 18:30-19:00**

Magic angle spinning NMR of paramagnetic proteins

Loren Andreas
Institut des Sciences Analytiques, Lyon, France

Abstract

The ubiquity of metal ions in biochemical and cellular processes, combined with the fact that many metal ions are paramagnetic by virtue of their unpaired electrons, make paramagnetic systems an important cl ass of targets for research in structural biology and related fields. Today, NMR spectroscopy plays a central role in the investigation of the structure and chemical properties of paramagnetic metalloproteins, in which the paramagnetic phenomena observed can be directly linked to electronic and molecular structure. A major step forwards in the study of proteins by solid-state NMR came with the advent of ultra-fast magic angle spinning (MAS) and ability to use 1H detection. Combined, these techniques have allowed investigators to observe nuclei in highly paramagnetic metalloproteins that previously were invisible, enabling quantitative site-specific measurement of a variety of long-range paramagnetic effects. This lecture covers aspects of solid-state NMR related to paramagnetic systems in biochemistry, with an emphasis on state-of-the-art methods and applications. In particular, the use of ultra-fast MAS and 1H-detection in paramagnetic metalloproteins is discussed. Current methodology allows the determination of the structure and dynamics of a wide range of biomolecules in remarkably short experimental times and requiring only a few milligrams of sample.

Date: 19 January 2016 **Time: 08:00-09:20**

Protein aggregation and amyloid formation associated with human diseases

Samir Maji
Indian institute of Technology Bombay, Mumbai, India

Abstract

Amyloids are highly order protein/peptide aggregates associated with numerous human disorders such as Alzheimers and Parkinsons. Amyloid fibrils are cross- β-sheet rich structure and formed by nucleation dependent polymerization reaction. Amyloids are cytotoxic and infectious in nature. Recent studies have suggested that the soluble oligomers are more cytotoxic and responsible for diseases rather than the mature fibrils. Therefore studying pathway(s) of protein aggregation and amyloid formation and developing structure-function relationship of toxic protein aggregates is important for therapeutic development against amyloid diseases.

Date: 19 January 2016 **Time: 09:40-10:40**

Dynamics of amyloid fibrils

Daniel Huster
Institut für Medizinische Physik und Biophysik, Universität Leipzig, Germany

Abstract

Amyloid fibrils are well ordered and highly structured assemblies of predominantly β-sheet secondary structures in a cross-β structural arrangement. Typically, these structures are perceived as highly rigid, but several recent studies indicate that these putatively stiff structures also reveal some dynamic features. In the talk, I will mostly dwell on the dynamical solid-state NMR methods introduced in the workshop before and discuss some recent findings on the dynamic nature of amyloid fibrils of natural amyloids, biologically important mutations as well as rationally designed fibrils. Analysis of the fibril dynamics represents a tool for the understanding of the packing properties and organization of amyloid fibrils.

Date: 19 January 2016 **Time: 11:00-12:00**

DNP of Proteins

Hartmut Oschkinat
Leibniz-Institut für Molekulare Pharmakologie, Berlin, Germany

Abstract

Date: 19 January 2016 **Time: 12:00-13:00**