Conference Program (as of 14-April-2022)

Refer to Online Planner & Mobile App for UPDATED PROGRAM.
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This Conference Program is subject to change. It is intended to serve as a reference guide only.

The online planner and mobile app tools for registered attendees will be updated regularly as changes occur.

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63rd ENC PROGRAM (as of 14-April-2022)

Refer to Online Planner & Mobile App for UPDATED PROGRAM. This file is general reference only.

SATURDAY, APRIL 23, 2022

IVAN Users' Meeting, Windsong
08:00-17:00

08:00-17:00 **Additional information and free RSVP pending**

BRUKER Workshop, Grand H
13:00-16:15

13:00-16:15 **Description and free RSVP here**

SUNDAY, APRIL 24, 2022

BRUKER Symposium, Grand DE
08:30-12:00

08:30-12:00 **Description and free RSVP here**

JEOL Mini-Symposium, Grand F
09:00-12:00

09:00-12:00 **Additional information and free RSVP pending**

National MagLab STEM Event – Kids & Family Science Activities
12:00-14:00, Oak Terrace (across from Pool)

Interactive demos and hands-on magnet activities geared toward K-8 students, but family members of all ages are welcome to join & interact. For additional information, check out the National MagLab's 'Magnet Academy' website.

The National High Magnetic Field Laboratory is the world's largest and highest-powered magnet facility. Located at Florida State University, the University of Florida and Los Alamos National Laboratory, the interdisciplinary National MagLab hosts scientists from around the world to perform basic research in high magnetic fields, advancing our understanding of materials, energy and life. The lab is funded by the National Science Foundation (DMR-1644779) and the state of Florida. For more information, visit us online at nationalmaglab.org or follow us on Facebook, Twitter, Instagram and Pinterest at NationalMagLab.

YOUNG SCIENTISTS' SYMPOSIUM (YSS)
13:30-16:00, Grand DE

Rieko Ishima and Len Mueller, presiding - Seven talk selected from young scientist poster presentations (Mon-Thurs)

YSS 13:30, **Georges Menzildjian** (CRMN) *TinyPol-like Biradicals for Highly Efficient DNP MAS NMR at High Magnetic Fields and Fast MAS*		YSS 14:40, **Monica Bastawrous** (Univ of Toronto) *Application of a 1mm Micro-coil Flow Probe with External Lock for In-Vivo NMR Metabolomics of Daphnia Neonates and Dormant Eggs*
YSS 13:50, **Kara Anazia** (University of Florida) *New Insights into the Structure-Function Relationships of G Proteins Containing Disease-Associated Mutations*		YSS 15:00, **Wes Pawloski** (Univ of Marland College Park) *NMR as a Tool to Measure Activation of Ubiquitin and Ubiquitin-like Proteins by E1 Enzymes*
YSS 14:10, **Rhythm Shukla** (Utrecht University) *A new antibiotic forms fibrils to destroy the bacterial membrane*		YSS 15:20, **Rittik Ghosh** (UC Riverside) *Active site chemistry in a 206 kDa PLP enzyme enabled by DNP enhanced Solid State NMR Spectroscopy*
TEN-MINUTE BREAK, 14:30 – 14:40		YSS 15:40, **Isaiah Adelabu** (Wayne State University) *Order-unity 13C Hyperpolarization of a-ketocarboxylates in Under 1 minute via SABRESHEATH: [1-13C]pyruvate, [1-13C]alpha-ketoglutarate and [1-13C]ketoisocaproate*

WELCOME RECEPTION with Exhibit Booths
16:00-18:00, Portico (foyer) with exhibit booths

OPTIONAL Vendor Hospitality Suites (following reception)

MONDAY, APRIL 25, 2022

EARLY AM: SPIN DYNAMICS (plenary session)
07:00-08:00

07:00-08:00 **Simulation Tutorial** Part 1 of 5: **BASICS**

Presenter: Ilya Kuprov

MOA: Laukien Prize Session (plenary session)
09:00-10:10, Grand DE
Joanna R. Long presiding

COFFEE BREAK with Exhibit Booths
10:10-10:45

MOC: Biomolecular Solutions 1: New Windows into Mechanism (parallel session)
10:45-12:35, Grand DE
Rieko Ishima presiding

10:45-11:10 **Monitoring "sulfur" sites via 77Se triple resonance experiments – and some instrumental gymnastics**

Presenting Author: R. Andrew Byrd

Janusz Koscielniak (Leidos Biomedical Research); Jess Li (National Cancer Institute); Deepak Sali (NHLBI/NIH); Rolf Swenson (NHLBI/NIH); Clemens Anklin (Bruker BioSpin); Sharon Rozovsky (Univ. Delaware); R. Andrew Byrd (National Cancer Institute)

NMR has been applied to virtually all sites within proteins and biomolecules; however, the observation of sulfur sites remains very challenging. Recent work from the Rozovsky lab has examined 77Se as a replacement for sulfur and applied 77Se NMR in the solid state, as well as some solution studies. Despite being a spin-1/2 nuclide, there have been rather limited studies of 77Se, and the ability to use 1H-indirect detection has been sparse. We will illustrate approaches using double-labeling of 13C and 77Se in proteins that enable more sensitive triple-resonance schemes. These methods also require specialized hardware and decoupling schemes, which we have developed and will be discussed.

11:10-11:30 **The Extended Hadamard Transform for Sensitivity-Enhanced NMR Experiments Among Labile 1Hs: Applications to SARS-CoV-2-Derived RNAs Detecting Correlations in High Dimensional 1GHz NMR**

Presenting Author: Jihyun Kim

Jihyun Kim (Weizmann Institute of Science); Mihajlo Novakovic (Weizmann Institute of Science); Tassilo Grün (Weizmann Institute of Science); Sundaresan Jayanthi (Indian Institute of Space Science and Technology); Adonis Lupulescu ("Horia Hulubei" National Institute for Physics and Nuclear Engineering IFIN-HH); Eriks Kupce (Bruker UK Ltd); Klara Mertinkus (University of Frankfurt); Andreas Oxenfarth (University of Frankfurt); Harald Schwalbe (University of Frankfurt); Lucio Frydman (Weizmann Institute of Science)

Hadamard saturation transfer can significantly increase the sensitivity of NOE correlations from labile protons in various biomolecules. However, this scheme generates artifacts when attempting correlations within a pool of labile protons. This limits the application of this method to measure, for example, imino-imino correlations in nucleic acids containing valuable information about base pairing and secondary structures. This study investigates the origin of these artifacts and proposes a way to record artifact-free correlations between labile sites and between labile and non-labile sites. This is demonstrated with 1GHz 2D NOESY-like correlations measured on RNA fragments derived from the SARS-CoV-2 virus, enhanced by an order of magnitude. Similar sensitivity gains can be observed when combining this scheme with 3D/4D NOESY/HSQC-type experiments.

MONDAY, APRIL 25, 2022 - continued

11:30-11:50 **How do Chaperones Achieve Client Specificity? Insights from the Mitochondrial Membrane-Protein Chaperones**

Presenting Author: Iva Sučec

Iva Sucec (Institut de Biologie Structurale); Katharina Weinhäupl (Universidade do Portoo, I3S - Instituto de Investigacao e Inovacao em Saude); Yong Wang (Structural Biology and NMR Laboratory, the Linderstrøm-Lang Centre for Protein Science, Department of Biology, University of Copenhagen); Ons Dakhlaoui (Univ. Grenoble Alpes, CEA, CNRS, IRIG, MEM); Tobias Jores (Department of Genome Sciences, University of Washington); Doriane Costa (Univ. Grenoble Alpes, CEA, CNRS, Institut de Biologie Structurale); Audrey Hessel (Institute of Pharmacology and Structural Biology, University of Toulouse, CNRS); Martha Brennich (Bruker Optics); Doron Rapaport (Interfaculty Institute of Biochemistry, University of Tübingen); Kresten Lindorff-Larsen (Structural Biology and NMR Laboratory, the Linderstrøm-Lang Centre for Protein Science, Department of Biology, University of Copenhagen); Beate Bersch (CNRS, Institut de Biologie Structurale); Paul Schanda (Institute of Science and Technology Austria)

The majority of mitochondrial proteins are synthesized in the cytosol and transported through the outer membrane in a post-translational manner. Two homologous intermembrane space (IMS) holdase chaperones, called small TIM chaperones, transport highly-hydrophobic membrane proteins. They bind different clients with different affinity, but the mode of binding and structural basis was unknown. Using solution NMR, biophysical, biochemical and molecular modeling approaches, we revealed that client specificity is achieved by a balance of hydrophobic and hydrophilic interactions of the chaperones with their client proteins. The hydrophobic interactions ensure some degree of promiscuity while the client selectivity between TM9.10 and TIM8.13 chaperone is polar-interaction driven, involving a distinct chaperone binding site.

11:50-12:10 **Dynamic, Real-Time Perspectives of Silicifying Peptide R5**

Presenting Author: Fanny Kozak

Fanny Kozak (University of Vienna/Institute of Biological Chemistry); Daniela Reichinger (University of Vienna/ Institute of Biological Chemistry); Ludovica Epasto (University of Vienna); Christian Friedrich Wilhelm Becker (University of Vienna/Institute of Biological Chemistry); Dennis Kurzbach (University of Vienna/Institute of Biological Chemistry)

Biomineralization processes such as bone graft and silica shell formation in diatoms remain incompletely understood. Still, their understanding at atom and residue resolution is essential to control the process and endow the silica with certain properties. Herein, we report a new, multidimensional perspective on the silica precipitation processes mediated by Silaffine mimetic peptide R5. By integrating state-of-the-art, high-resolution liquid-state NMR techniques and molecular dynamics simulations, residues most affected were identified. To further observe the developments directly as they unfold, 1H-15N amide backbone SOFAST-HMQC were recorded every two minutes, resulting in residue resolution, time-resolved kinetics for the accessible amino acids. Grave differences have been observed depending on the concentration of phosphate ions, which are central for the precipitation event.

12:10-12:35 **IDPs Under Physiological Conditions: Proline cis/trans Isomers, the Effect of Phosphorylation - Revealed by 1Hα-detected NMR Methods**

Presenting Author: Andrea Bodor

Fanni Sebák (ELTE Eötvös Loránd University, Institute of Chemistry); Péter Ecsédi (ELTE Eötvös Loránd University, Department of Biochemistry); Wolfgang Bermel (Bruker Biospin GmbH); Burkhard Luy (Karlsruher Institut für Technologie - KIT); László Nyitray (ELTE Eötvös Loránd University); Andrea Bodor (ELTE Eötvös Loránd University)

Characterization of intrinsically disordered proteins (IDPs) is most relevant at physiological conditions. Proline residues are frequently abundant in IDPs and the cis/trans isomerization of these environments contributes to the conformational heterogeneity. We developed novel 1Hα-detected approaches to characterize both high and low populated species and to unambiguously report on the cis or trans proline isomeric form. Using p53TAD1-60 with 10 proline environments we demonstrate that the new measurements are fully resolved, and sensitive enough to identify minor forms in 4-15% amounts. It is also possible to monitor the response of post-translational modifications on the cis/trans–proline equilibrium. Regarding the varying amounts of minor forms, we analysed available literature data on how the amino acid type affects the cis/trans-proline distribution.

MONDAY, APRIL 25, 2022 - continued

MOD: Theory and Computation 1: From Solids Tensors to Quantum Chemistry and Back

(parallel session)
10:45-12:35, Grand F
David Rovnyak presiding

10:45-11:10 **Drugs and drug-polymer formulations on the dissecting table –Insights through NMR spectroscopy and complementary tools in the solid state and in biorelevant solutions**

Presenting Author: Ann-Christin Pöppler

Sebastian Endres (University of Wuerzburg); Marvin Grüne (University of Wuerzburg); Ann-Christin Pöppler (University of Wuerzburg)

Both the assembly of amorphous drug-polymer micellar formulations and their behaviour in biological environments are very diverse.

Solid-state NMR and quantum chemical calculations can reinforce each other to obtain insights into the molecular level assembly of paclitaxel as well as loaded poly-(2-oxazoline) polymer micelles, for which we could identify a new crystallographic anhydrous phase and a loading dependent structural model, respectively.

Subsequently, we follow drug-polymer formulations in biorelevant media such as fed-state simulated intestinal fluids (FeSSIF). NMR spectroscopic analysis in solution complemented by quantum chemical calculation and cryo-TEM reveals a rich concentration and composition dependent colloidal aggregation behaviour underlining that a screening of individual polymers or drugs would often not be sufficient.

11:10-11:30 **Ab Initio Crystal Structure Prediction: NMR Crystallography Using Quadrupolar Nuclei (CSP-QNMRX)**

Presenting Author: Austin A. Peach

Austin Peach (Florida State University); Carl H. Fleischer (Florida State University); Kirill Levin (Université de Sherbrooke); Sean T. Holmes (Florida State University); Robert W. Schurko (Florida State University)

The use of electric field gradient (EFG) tensors of quadrupolar nuclei (e.g., 14N, 17O, 23Na, etc.) derived from experiment and theory for crystal structure prediction (CSP) using an NMR crystallographic approach (QNMRX) has largely gone unexplored. EFG tensors at the quadrupolar nuclides are extremely sensitive to even the most subtle differences or changes in structure; hence, they are excellent probes of local atomic environments and structural features. Herein, we demonstrate the utility of 35Cl EFG tensors in a new CSP-QNMRX protocol for the prediction and refinement of crystal structures of organic HCl salts. Five simple rigid HCl salts are used to benchmark key metrics and filters, which are extended to several “blind” tests on organic HCl salts with unknown structures.

11:30-11:50 **Machine Learning and Data-Driven Methods in Solid-State NMR: Unlocking New Tools for NMR Crystallography**

Presenting Author: Manuel Cordova

Manuel Cordova (EPFL); Lyndon Emsley (EPFL)

Solid-state NMR, in combination with computational methods such as density functional theory chemical shift computation and crystal structure prediction (CSP) procedures, has proven able to determine the crystal structure of organic solids. The recent introduction of ShiftML, a machine learning model of chemical shifts, has enabled both a substantial acceleration of chemical shift computations and access to shifts of large ensembles of large structures. Here, we present new tools derived from ShiftML that can be used to accelerate and improve the structure determination of materials by assisting the interpretation of NMR spectra, acting as a driving force in crystal structure prediction protocols, or allowing direct comparison between large structural models and experiment to determine the structure of amorphous compounds.

MONDAY, APRIL 25, 2022 - continued

11:50-12:10 **^{27}Al NMR quadrupolar and chemical shielding tensors benchmarking with DFT: prediction of quadrupolar coupling constants (C_Q) from simple local geometry and elemental properties**

Presenting Author: He Sun

He Sun (Washington University in St.louis); Michael E. West (Department of Chemistry, Washington University in St.louis); Shyam Dwaraknath (Lawrence Berkeley National Laboratory); Handong Lin (Department of Materials Science and Engineering, UC Berkeley); Kristin A. Persson (Department of Materials Science and Engineering, UC Berkeley); Hayes E. Sophia (Department of Chemistry, Washington University)

We developed a simple machine learning ("random-forest") model based on local structural motifs and elemental properties to predict tensor values for the quadrupolar nucleus ^{27}Al. This catalog permits a rapid assignment of species before validating with first-principles calculations. Prediction of the quadrupolar coupling constant (C_Q) for aluminum-containing crystalline materials was yielded good agreement when comparing to DFT-computed values (RMSE of 0.61 MHz; R^2=0.98). Simple geometric features dominated the predictive accuracy. The model was trained on a computational dataset of 1,562 NMR spectra using VASP. The dataset was benchmarked against both CASTEP and 105 experimental values compiled from literature, which validated the accuracy of the DFT-NMR parameters for ^{27}Al chemical shielding and EFG tensors.

12:10-12:35 **Improving the Accuracy of Calculated NMR Parameters in Solids**

Presenting Author: Jonathan Yates

Jonathan Yates (University of Oxford)

For a number of years it has been possible to predict NMR tensors for both isolated molecules and extended solids using quantum chemistry techniques - often to a useful level of accuracy. With each generation of computer processors the time to complete calculations decreases. This means we can treat increasing complicated structures or consider ensemble models for either configurational or dynamic disorder. We might say that we are increasing the accuracy of the simulation by considering more realistic models. However, in this talk I will consider how we might increase the fundamental accuracy of these simulations; focusing on solids I will discuss prospects for more exact solutions of the Schroedinger equation, and the effects of relativity on NMR parameters.

LUNCH-ON-YOUR-OWN
12:35-14:00

POSTER SESSION
14:00-15:45, Regency Hall
See Poster Listings at end of document

MOE: Biomolecular Solids 1: Atomic Level Insights into Complex Architectures (parallel session)
16:00-17:50, Grand DE
Leonard Mueller presiding

16:00-16:25 **Fluorine MAS NMR and DNP of Molecules Small and Large: Spin it Fast!**

Presenting Author: Tatyana Polenova

Tatyana Polenova (University of Delaware); Changmiao Guo (University of Delaware); Manman Lu (University of Pittsburgh School of Medicine); Brent Runge (University of Delaware); Roman Zadorozhnyi (University of Delaware); Caitlin M. Quinn (University of Delaware); Sucharita Sarkar (Graduate student); Gal Porat-Dahlerbruch (University of Delaware); Jochem Struppe (Bruker Corporation); Ivan Sergeyev (Bruker Corporation); Angela M. Gronenborn (University of Pittsburgh School of Medicine)

Recent methodological advances in 19F MAS NMR and DNP will be discussed, with applications in small-molecule pharmaceuticals, pharmaceutical formulations, microcrystalline proteins and large biological assemblies. It will be demonstrated that high MAS frequencies (40-100 kHz) are advantageous for the homo- and heteronuclear correlation experiments, the measurement of accurate interfluorine distances, and recoupling of 19F chemical shift anisotropy. With remarkably high, up to 100-fold, signal enhancements were observed in 19F DNP MAS NMR spectra of HIV-1 capsid protein assemblies, it was possible to record 2D 19F-13C HETCOR spectra. These spectra contain long-range intra- and intermolecular correlations, which are not easily accessible in conventional experiments without DNP.

MONDAY, APRIL 25, 2022 - continued

16:25-16:45 **High-Field and DNP Solid-State NMR to Probe Whole Algae Cells Glycan Composition and Cell-Wall Glycoprotein Architecture**

Presenting Author: Alexandre Poulhazan

Alexandre Poulhazan (Université du Québec à Montréal); Alexandre A. Arnold (Université du Québec à Montréal); Frederic Mentink-Vigier (National High Magnetic Field Laboratory); Dror E. Warschawski (Laboratoire des Biomolécules, LBM, CNRS UMR 7203, École normale supérieure, PSL University); Tuo Wang (Department of Chemistry, Louisiana State University); Isabelle Marcotte (Université du Québec à Montréal)

We report high-field and MAS-DNP ssNMR experiments to describe, with atomic resolution, the native cell wall architecture and whole cell glycan composition of two microalgae, Chlamydomonas reinhardtii and Parachlorella beijerinckii. To do so, several one- and multi-dimensional experiments, such as ^{13}C-^{13}C INADEQUATE or water-edited DARR, allowed investigating the role of glycans in the structure, hydration, and flexibility of polysaccharides or glycoproteins. The identification of rigid and hydrated regions allowed us to propose a structural model of cell-wall glycoproteins and whole-cell glycans. Altogether, this work shows that ssNMR can contribute to our understanding of cell surface protection and the role of protein glycosylation.

16:45-17:05 **Proton Positions, Positional Uncertainties, and Role in Inhibition of the Tryptophan Synthase α-Aminoacrylate Intermediate**

Presenting Author: Jacob B. Holmes

Jacob B. Holmes (Department of Chemistry, University of California, Riverside); Rittik K. Ghosh (Department of Biochemistry, University of California, Riverside); Viktoriia Liu (Department of Chemistry, University of California, Riverside); Jennifer A. Romero (Department of Chemistry, University of California, Riverside); David Amarasinghe (Department of Chemistry, University of California, Riverside); Leonard J. Mueller (Department of Chemistry, University of California, Riverside)

NMR-crystallography is used to define the active site protonation states in the tryptophan synthase α-aminoacrylate intermediate. Confidence in the identification of the experimental structures is assigned using reduced chi-squared and Bayesian probability analysis, while positional uncertainties on the atom coordinates are determined using the low-T MD method of Hofstetter and Emsley (JACS 2017) extended to our cluster approach. Average positional RMSD of 0.11-0.17 Å are found, similar to those in molecular organic crystals and 6.5 times smaller than those from protein X-ray crystal structures. These results suggest that positional uncertainties in NMR crystallography are independent of molecular size. For the α-aminoacrylate, this approach allows the identification of the initial binding pocket for the eliminated water and the mechanism of inhibition.

17:05-17:25 **MAS NMR View of Melanin Pigment Deposition in Fungal Cell Walls: Laying the Foundation, Greasing the Wheels**

Presenting Author: Ruth E Stark

Christine Chrissian (City University of New York); Emma Camacho (Johns Hopkins Bloomberg School of Public Health); John E. Kelly (City University of New York); Hsin Wang (City University of New York); Arturo Casadevall (Johns Hopkins Bloomberg School of Public Health); Ruth Stark (City University of New York)

Despite the association of melanin pigments with fungal virulence in humans and inspiration for the design of nanomaterials used in electron transport and drug delivery, the molecular factors that control their development and deposition remain poorly understood because they are insoluble, amorphous, and chemically heterogeneous. A suite of 1D and 2D solid-state MAS NMR experiments, (DARR, TEDOR, DP J-INADEQUATE, INEPT-HETCOR) has been used to examine melanized cell walls (‘ghosts’) and whole Cryptococcus fungal cells, focusing on the cell-wall composition, scaffold flexibility, and lipid classes that can impact pigment deposition under metabolic stress conditions. Together with TEM and cell-based assays, these studies of macromolecular architecture for a biological composite offer new insights into melanin’s functional impact on fungal pathogenicity.

MONDAY, APRIL 25, 2022 - continued

17:25-17:50 **The Functional Interplay Between Lipids, Channels, Receptors, and Biological Assemblies as Viewed by Solid-state NMR**

Presenting Author: Benjamin James Wylie

Benjamin Wylie (Texas Tech University); Collin Griffin Borcik (Texas Tech University); Reza Amani (Department of chemistry and biochemistry, Texas Tech University); Evan Jamaal van Aalst (Texas Tech University); Maryam Yekefallah (Texas Tech University); Isaac R. Eason (Texas Tech University); Carver Rasberry (Texas Tech University); Jun Jang (Texas Tech University)

SSNMR studies of the KirBac1.1 K+ channel and CCR3 GPCR will be described. We determined the mechanism and conformational changes associated with KirBac1.1 activation by anionic lipids and solved the structure of the activated and inactivated states. This established a new set of water-edited restraints for simulated annealing. We then reveal a persistent cholesterol dimer occupies a conserved Kir channel cholesterol recognition site. Next, current work on the CCR3 C-C motif chemokine GPCR will be presented. We discovered that the affinity between CCR3 and the CCL11 chemokine and the GTPase activity of CCR3-activated Gαi3 are both cholesterol dose-dependent. These observations are consistent with molecular dynamics simulations. SSNMR investigation of these functional mechanisms confirms CCR3 fold is conformationally selected by cholesterol.

MOF: In-Vivo MRI (In-Vivo / Ex-Vivo 1) (parallel session)
16:00-17:50, Grand F
Malgorzata 'Gosia' Marjanska presiding

16:00-16:25 **A Statistical Framework for Magnetic Resonance Fingerprinting: Optimized Encoding and Decoding**

Presenting Author: Bo Zhao

Bo Zhao (University of Texas at Austin)

Magnetic Resonance Fingerprinting (MRF) is a transient-state quantitative MRI paradigm, which enables rapid acquisitions of multiple MR tissue parameter maps in a single imaging experiment. Despite its novel concept, MRF has several inherent limitations, which constrains its utility. In this talk, I will present a novel statistical framework that we have recently introduced to optimize the encoding and decoding processes of MRF. On the decoding side, we have shown the sub-optimality of the pattern-matching based MRF reconstruction and have introduced several principled statistical approaches to estimate MR tissue parameters from k-space data, which significantly improves the reconstruction accuracy. On the encoding side, we have optimized MRF acquisition parameters based on the estimation-theoretic metrics to improve the acquisition SNR efficiency.

16:25-16:45 **Quantifying Murine Tracheal Dynamics Using Retrospectively Gated Ultra-short Echo-time MRI**

Presenting Author: Qing Wang

Qing Wang (Cincinnati Children's Hospital Medical Center); Zackary Cleveland (Cincinnati Children's Hospital Medical Center); Elizabeth Fugate (Cincinnati Children's Hospital Medical Center)

Tracheomalacia is the collapse of the primary conducting airway (trachea) during exhalation and results from congenital malformation of or damage to the normally rigid tracheal cartilage. Tracheomalacia occurs as a co-morbidity in a range of disorders, and for example, may play a pathological role in cystic fibrosis (CF), a fatal genetic disease involving progressive airway damage. Recent evidence showed congenital airway malformation occurs even without airway injury in transgenic CF mouse models, creating an unmet need to study pathological upper airway dynamics in small animal. Here we report an MRI acquisition protocol based on radial, ultra-short echo-time (UTE) with retrospective gating that yields sufficiently high resolution to measure airway caliber changes during normal tidal breathing in mice.

16:45-17:05 **The Influence of Systematic and Statistical Errors on the DTI Metrics and Tractography of Rat's Brain White Matter**

Presenting Author: Weronika Mazur

Weronika Mazur (AGH University of Science and Technology); Anna Stefańska-Bernatowicz (AGH University of Science and Technology); Artur T. Krzyżak (AGH University of Science and Technology)

The study shows the joint effect of noise and systematic errors (SE) correction with the application of B-matrix spatial distribution (BSD) method on the DTI metrics of a rat's brain white matter (corpus callosum, external capsule, cingulum). BSD with noise reduction applying Gaussian filter significantly reduced eigenvalues branching error and exhibited the best performance among tested methods in isotropic phantom. Results from brain show, that at SNR=34-51 and after BSD application, relative differences between erroneous and corrected data total -12%, 6%, 65%, 2% and -26% for λ1, λ2, λ3, MD and FA, respectively. The joint effect of BSD and noise reduction on DTI tractography yielded a tremendous improvement in fiber tracts density with anatomically correct directions.

MONDAY, APRIL 25, 2022 - continued

17:05-17:25 **Novel Dynamic Nuclear Polarization 13C Probes Reveal Real-time Aberrant Cancer Metabolisms In Vivo**

Presenting Author: Kazutoshi Yamamoto

Kazutoshi Yamamoto (National Institutes of Health / National Cancer Institute); Yohei Kondo (The University of Tokyo); Yutaro Saito (The University of Tokyo); Peter L. Choyke (National Institutes of Health / National Cancer Institute); James B. Mitchell (National Institutes of Health / National Cancer Institute); Rolf E. Swenson (National Institutes of Health / National Heart, Lung, and Blood Institute); Shinsuke Sando (The University of Tokyo); Murali C. Krishna (National Institutes of Health / National Cancer Institute)

Dissolution Dynamic Nuclear Polarization (dDNP) is an emerging technique to detect site-specific physiology and enzymatic activities in in vivo conditions. While drastic dDNP sensitivity enhancements are advantageous for non-invasive real-time measurements, the limited number of applicable probes with longer T1 relaxation times continues to be a major drawback particularly for in vivo use. In this presentation, we will demonstrate the feasibility of newly synthesized hyperpolarized 13C probes including N-Acetyl Cysteine, Glutathione, Aminopeptidase N probes, gamma-glutamyl transferase probes, beta-Casomorphin probes, alpha-ketoglutaric acid and its derivatives. This wide variety of probes effectively reveals unique cancer metabolisms and specific enzymatic activities related to progression of cancer, including redox status and clinical markers. Advantages and limitations of our novel probes will also be discussed.

17:25-17:50 **MR Fingerprinting and Optimization**

Presenting Author: Dan Ma

Dan Ma (Case Western Reserve University)

Magnetic Resonance Fingerprinting (MRF) is a quantitative imaging method to simultaneously provide multiple tissue properties such as T1 and T2 relaxations times from a single scan. With its high scan efficiency and reproducibility, MRF has been applied to various clinical studies for disease detection and characterization, such as brain tumors, epilepsy, and multiple sclerosis. MRF pulse optimization has been challenging because an MRF sequence has thousands of tunable parameters, which can be chosen to maximize precision and minimize scan time. In this talk, I will introduce the MR Fingerprinting concept, various sequence optimization considerations, and clinical applications.

Vendor Hospitality Suites
From 18:30

AMMRL Meeting
19:00-20:30
Marc ter Horst presiding

TUESDAY, APRIL 26, 2022

EARLY AM: SPIN DYNAMICS (plenary session)
07:00-08:00

07:00-08:00 **Simulation Tutorial** Part 2 of 5: **LIQUIDS**

Presenter: Ilya Kuprov

TOA: Instrumentation, Hyperpolarization, & Eclectica 1: Hyperpolarization and NMR Spectroscopy at Low Field (parallel session)
08:30-10:20, Grand DE
Carl Michal presiding

08:30-08:55 **Exploring Parahydrogen Hyperpolarisation using Low-field NMR Instrumentation**

Presenting Author: Meghan E. Halse

Meghan Halse (University of York)

Signal amplification by reversible exchange (SABRE) is a catalytic method for transferring polarisation from parahydrogen to a target analyte in solution via a reversible exchange reaction. SABRE transfer is optimised in a very weak magnetic field on the order of 0 - 10 mT. In a standard SABRE experiment, polarisation transfer is carried out in a weak magnetic field and then the sample is transported to the NMR spectrometer for detection. This two-stage approach makes it very difficult to directly observe the dynamics of the SABRE transfer. In this presentation an in situ approach will be introduced that uses field cycling and Earth's field NMR detection to explore the dynamics of SABRE directly.

08:55-09:15 **A Hybrid LAC-INEPT Approach for Highly Efficient Spin Order Transfer in Side-Arm Hydrogenation with Parahydrogen**

Presenting Author: Maria-Jose Ferrer

Maria-Jose Ferrer (University of Florida); Clifford R. Bowers (University of Florida)

Spin-order transfer (SOT) after side-arm-hydrogenation (SAH) with parahydrogen is a promising approach to the hyperpolarization of 13C in carbonyl-containing metabolites. We introduce a hybrid SOT method for hyperpolarization of 13C in acetate and pyruvate after SAH with parahydrogen. The rapid adiabatic passage through a level anti-crossing (LAC) resulting from homonuclear proton-proton couplings and Zeeman interactions can spontaneously convert the parahydrogen singlet-order into net alignment of a third initially unpolarized proton in the side-arm with high efficiency. The required combination of spin Hamiltonian parameters to optimize this process has been elucidated. This hybrid SOT via rapid adiabatic passage by proton-proton LAC combined with a coherence transfer pulse sequence at high field can provide significantly higher 13C hyperpolarization than previously reported SOT-techniques.

09:15-09:35 **Non-Intuitive Modulated Fields Improve Polarization Transfer in SABRE**

Presenting Author: Shannon L Eriksson

Shannon Eriksson (Duke University); Warren S. Warren (Duke University)

Signal Amplification By Reversible Exchange (SABRE) methods have traditionally been constructed around either matching a low field to energy level differences or using radiofrequency pulses to shift levels into resonance, ideas that are rooted in conventional magnetic resonance techniques and avoided crossings. However, using our physically accurate numerical simulations we have been able investigate field conditions outside of this theoretical regime to find non-intuitive conditions which better optimize polarization transfer. We demonstrate that oscillations in the leading field, which never bring the system to traditional SABRE-SHEATH polarization transfer conditions yields a 3-fold improvement in polarization. Additionally, we show that application of fields along multiple axes at low field can lead to more targeted and optimal polarization transfer.

TUESDAY, APRIL 26, 2022 - continued

09:35-09:55 **Measurement of Fast Electron Spin Relaxation Times at Ultra-low Fields**

Presenting Author: Xueyan Tang

XUEYAN TANG (University of Minnesota, Center for Magnetic Resonance Research); Steven Suddarth (University of Minnesota, Center for Magnetic Resonance Research); Saurin Kantesaria (University of Minnesota, Center for Magnetic Resonance Research); Michael Garwood (University of Minnesota, Center for Magnetic Resonance Research)

The relaxation time of an electron spin is important for several research endeavors, including the design of MRI contrast agents and iron-oxide nanoparticles for MRI, hyperthermia, and magnetic particle imaging. However, its value is field-dependent, and the measurement of fast spin relaxation times (< nanoseconds) at low fields (< 50 Gauss) is challenging due to poor signal-noise ratio. We built a broad-band, portable EPR system that operates below 10 Gauss. This system combines longitudinal detection with fictitious field modulation, which enables simultaneous transmit and receive with -80 dB isolation. By using a new equation derived in our previous work, here we show that fast spin relaxation times can be calculated from the peak shift in the frequency-swept EPR spectra.

09:55-10:20 **No Resolution, No Problem: NMR Spectroscopy at Low Magnetic Fields**

Presenting Author: Stephen J. DeVience

Stephen DeVience (Scalar Magnetics)

Low-field NMR has historically been relegated to specialized uses such as educational MRI demonstrations, rock-core analysis, and relaxometry. However, with well-developed methods of J-coupling spectroscopy for heteronuclear spin systems and new pulse sequences such as spin-lock induced crossing (SLIC) for homonuclear systems, many classes of molecules can also be studied spectroscopically even at low fields. I will discuss recent advances in the field and results from experiments using a 6.5 mT instrument, focusing in particular on applications where J-coupling spectroscopy can have advantages over high-field techniques, such as measurements of weak spin-spin coupling and slow chemical exchange.

TOB: Materials 1: Batteries, Catalysts, and Paramagnetic Dopants (parallel session)
08:30-10:20, Grand F
Danielle Laurencin presiding

08:30-08:55 **Studies of Lithium-ion Dynamics and Structural Changes in LiFeV2O7 by Solid-State NMR**

Presenting Author: Gillian Goward

Taiana L.E. Pereira (McMaster University); Kevin J. Sanders (McMaster University); James F. Britten (McMaster University); Dany Carlier-Larregaray (Université de Bordeaux); Gillian Goward (McMaster University)

7Li solid-state nuclear magnetic resonance (ssNMR) was used to characterize the structure and dynamics of lithium rates in monoclinic LiFeV2O7. DFT calculations are utilized to assign the chemical shifts observed in the experimental spectrum of the as-synthesized material. Ex-situ 7Li magic angle spinning (MAS) ssNMR is used to track structural changes of the LiFeV2O7 electrode during electrochemical cycling. A new lithium arrangement was observed during the lithium insertion process which occurs at a similar point of the electrochemical process as a notable increase of the lithium-ion dynamics as observed by 2D EXSY and 7Li selective inversion experiments. The activation energy increases as a function of the lithiation, suggesting that the lithium vacancies play a significant role in the current dynamics.

08:55-09:15 **Dipolar Heteronuclear Correlation Solid-State NMR Spectroscopy Experiments Between Quadrupolar Nuclei**

Presenting Author: Rick W. Dorn

Rick Dorn (Iowa State University; US DOE Ames Laboratory); Ivan Hung (National High Magnetic Field Laboratory); Lesli O. Mark (University of Wisconsin - Madison); Alexander L. Paterson (US DOE Ames Laboratory); Peter L. Gor'kov (National High Magnetic Field Laboratory); Melissa C. Cendejas (University of Wisconsin - Madison); Ive Hermans (University of Wisconsin - Madison); Zhehong Gan (National High Magnetic Field Laboratory); Aaron J. Rossini (Iowa State University; US DOE Ames Laboratory)

There is great interest in the development of NMR experiments that can probe structural connectivity between quadrupolar spins, which make up 73% of all NMR active nuclei. Here we demonstrate pulse sequences for magic-angle spinning (MAS) 11B-17O Rotational-Echo Saturation-Pulse-DOuble Resonance (RESPDOR) and 2D Dipolar Heteronuclear Multiple Quantum Correlation (D-HMQC). In these pulse sequences, Rotational-Echo DOuble-Resonance (REDOR) recoupling with central-transition selective pi-pulses were applied to either the 11B or 17O spins. Using these methods, we obtained 2D 11B{17O} D-HMQC NMR spectra of 17O-enriched boronic acids, a sodium borate glass and a heterogeneous catalyst consisting of amorphous boron oxide on carbon. The latter two examples illustrate the power of these experiments for structure determination of disordered materials.

TUESDAY, APRIL 26, 2022 - continued

09:15-09:35 **Monitoring T1e with PRE and what we can learn from it**

Presenting Author: Daniel Jardón-Álvarez

Daniel Jardón-Álvarez (Weizmann Institute); Tahel Malka (Weizmann Institute); Johan van Tol (Florida State University); Yishay Feldman (Wezimann Institute); Raanan Carmieli (Weizmann Institute); Michal Leskes (Weizmann Institute)

NMR properties of materials containing unpaired electrons are controlled by the electron spin relaxation properties. However, measuring electron relaxation can be challenging, particularly at fields and temperatures of interest for NMR spectroscopy and DNP. We employ nuclear paramagnetic relaxation enhancement to map the relaxation of the electron spins by taking advantage of the fact that the ratio of the nuclear relaxation times, T_1/T_2, is proportional to the square of T_{1e}. By measuring T_1/T_2 ratios for varying paramagnetic centers content and temperature we are able to (i) rationalize the presence and absence of signal quenching upon increasing paramagnetic doping and (ii) find a linear relation between the signal enhancement obtained in DNP from metal ions and the determined T_{1e}.

09:35-09:55 **Characterising Crossover in Operating Aqueous Organic Redox Flow Batteries Using In Situ NMR**

Presenting Author: Emma Jane Latchem

Emma Latchem (University of Cambridge); Thomas Kress (University of Cambridge); Evan Wenbo Zhao (Radboud University); Clare P. Grey (University of Cambridge); Peter A. A. Klusener (Shell Global Solutions International B.V.); R. Vasant Kumar (University of Cambridge); Alexander C. Forse (University of Cambridge)

Aqueous organic redox flow batteries (AORFBs) are prospective devices for long-duration energy storage, which is required to integrate more renewable energy sources onto the electricity grid. However, crossover of redox-active species through the separator membrane can lead to irreversible capacity fade in these systems, limiting their lifetime and economic viability. It has previously been demonstrated that in situ spectroscopic techniques are powerful tools for determining reaction mechanisms in redox flow batteries. Here, we explore how solid-state nuclear magnetic resonance (NMR) spectroscopy and solution-state in situ NMR can be used to elucidate how charging protocols and membrane structure-property relationships govern electrolyte crossover. Ultimately, these fundamental studies will improve our understanding of electrolyte crossover, so that improved separator membranes can be developed.

09:55-10:20 **Unfolding the Surface Ligands on Nanocrystals under the Multifaceted Probe of NMR**

Presenting Author: Xueqian Kong

Zhenfeng Pang (Zhejiang University); Weicheng Cao (Zhejiang University); Xiaoqi Zhou (Zhejiang University); Xueqian Kong (Zhejiang University)

Nanocrystals are an important class of materials which have shown great promise in next-generation optoelectronic devices, heterogeneous catalysis and biomedical tracers. The nanocrystals consist of an inorganic core and a layer organic surface ligands. While the inorganic core contributes to the optoelectronic properties, the surface ligands determine the surface structure and control the assembly processes. However, the research on surface ligands are hampered by their packing disorder and intrinsically weak interactions. In recent works, we applied a set of solid-state and solution-state NMR techniques and gained insights on the surface ligands in multiple aspects, including the surface chemical bonds, surface morphology, ligand-ligand interactions and ligand exchange processes.

COFFEE BREAK with Exhibit Booths
10:20-10:45

TOC: Biomolecular Solids 2: Probing Biomolecular Assemblies (parallel session)
10:45-12:35, Grand DE
Gaël De Paëpe presiding

10:45-11:10 **NMR spectroscopic investigations of aggregates involved in AL-amyloidosis**

Presenting Author: Bernd Reif

Bernd Reif (TU Muenchen)

We investigate the structural mechanism of antibody light chain amyloid formation. MAS solid-state NMR experiments yield information on the conformation of the amyloidogenic core and allow to probe interactions with small molecules that potentially interfere with the aggregation process. We focus on human protein sequences for which adipose tissue and heart samples from patients are available. We discuss the potential of seeding strategies to reproduce in vivo amyloid fibril structures for in vitro produced labeled protein.

TUESDAY, APRIL 26, 2022 - continued

11:10-11:30 **Structural Basis of HIV-1 Maturation Inhibitor Binding and Activity by Solid State MAS NMR**

Presenting Author: Sucharita Sarkar

Sucharita Sarkar (Graduate student); Kaneil K. Zadrozny (Laboratory Technician 2); Roman Zadorozhnyi (Graduate student); Ryan W. Russell (Graduate student); Caitlin M. Quinn (Assistant professor); Alex Kleinpeter (Postdoctoral fellow); Sherimay Ablan (Research biologist); Chaoyi Xu (Graduate student); Juan R. Perilla (Assistant professor); Eric O. Freed (Senior Investigator); Barbie K. Ganser-Pornillos (Assistant professor); Owen Pornillos (Associate professor); Angela M. Gronenborn (Professor); Tatyana Polenova (Professor)

HIV maturation inhibitors (MIs), such as bevirimat (BVM), interfere with the final step of proteolytic cleavage of the HIV-1 Gag polyprotein: the cleavage of spacer peptide 1 (SP1) from capsid protein (CA) C-terminal domain (CACTD). MI binds to and stabilizes CTD-SP1 junction. We investigated CACTD-SP1 assemblies with IP6 in the presence and absence of BVM by MAS NMR. We established a protocol to solve structures of isotope-labeled protein assemblies in complex with natural abundance inhibitors using a suite of HETCOR experiments with and without dREDOR filters. Our results reveal atomic details of MI binding to CACTD-SP1. We obtained 3050 unambiguous intra-protein distance restraints and determined atomic resolution structures of CACTD-SP1 assemblies with IP6 in the presence and absence of BVM.

11:30-11:50 **Selective 17O NMR Spectroscopy at Ultra-High Field Magnet (35.2 T) Reveals Multiple Highly Stable Waters in Hydrated Phospholipid Bilayers**

Presenting Author: Rongfu Zhang

Rongfu Zhang (Postdoc); Xinhua Peng (Professor); Tim A. Cross (Professor); Riqiang Fu (Research Faculty)

Understanding water dynamics and structure is an important topic in biological systems. Although NMR is a powerful tool for structural and dynamic studies, direct probing of interfacial water in hydrated phospholipids is challenging due to large population difference between bulk and interfacial water. We develop a novel 17O solid-state NMR technique in combination with an ultra-high field magnet (35.2 T) and 17O enriched water to directly probe the interfacial water molecules. By selectively suppressing the dominant bulk water signal, we observed two chemically and dynamically distinct water species in the headgroup region of hydrated DMPC lipid bilayers. These new NMR discoveries clearly indicate the existence of the interfacial water molecules that are relatively stable over the millisecond timescale.

11:50-12:10 **3D-Printed Centrifugal Sample Rotor Preparation Devices for >100 kHz MAS NMR**

Presenting Author: Thomas Osborn Popp

Tom Osborn Popp (Rutgers University); Jacqueline Perodeau (Rutgers University); Ashley Bernstein (Rutgers University); Andrew Nieuwkoop (Rutgers University)

The advent of magic angle spinning (MAS) rates >100 kHz has enabled the acquisition of 1H-detected biomolecular solid state NMR spectra with high resolution. However, challenges can arise when preparing the sample rotors for these experiments, due to the physical properties of biomolecular solids and the small dimensions (0.5 mm inner diameter) of the rotors. We have developed 3D-printable, centrifugal sample preparation devices for 0.7 mm rotors, as well as protocols for successfully maintaining the hydration state of the sample while capping the rotor. Using these methods, we can reproducibly fill 0.7 mm rotors with well-hydrated nanocrystalline protein or lipid membranes at high packing efficiencies. We demonstrate the efficacy of these protocols using 1H-detected solid state NMR at 105 kHz.

12:10-12:35 **Re-evaluating Polarization Agent Design for In-cell DNP Applications**

Presenting Author: Galia Debelouchina

Galia Debelouchina (UC San Diego)

In recent years, there has been considerable interest in the development of DNP-enhanced NMR approaches for structural studies in cellular environments. While these efforts have highlighted the promise of DNP for structural biology in cells, they have also underscored the limitations of current DNP polarization agents for efficient signal enhancement in the cellular interior. This includes issues such as polarization agent selectivity, stability and cell permeability. In this presentation, I will discuss my group's efforts to address these limitations and to design and optimize targetable polarization agents for DNP studies in complex biological environments, including bacterial and mammalian cells.

TUESDAY, APRIL 26, 2022 - continued

TOD: In-Vivo / Ex-Vivo 2 (parallel session)
10:45-12:10, Grand F
Malgorzata 'Gosia' Marjanska presiding

10:45-11:10 **Title Pending**

Presenting Author: Moritz Zaiss (Max-Planck Institute, Tuebingin)

11:10-11:30 **Towards Robust, High-Stability 31P Spectral Editing to Quantify NAD+ and NADH Using an M2S-Inspired Approach**

Presenting Author: David E. Korenchan

David Korenchan (New York University); Alexej Jerschow (New York University)

Accurate, robust quantification of NAD^+ and NADH *in vivo* can improve disease diagnosis and treatment. However, NMR spectral overlap of NAD^+, NADH and the alpha-phosphate of ATP makes quantification difficult. We report ^{31}P spectral editing sequences that utilize a component of the nuclear singlet M2S-S2M sequences to isolate NAD^+ or NADH while suppressing the other two resonances. The sequences have editing efficiency near unity and are robust to both B_0 inhomogeneity and transmitter offset over a large range. The approach likely shows high stability by obviating the need for spectral subtraction. Future work will include phantom MRI studies to evaluate NAD^+/NADH ratio quantification via ^{31}P imaging.

11:30-11:50 **The Inside Story: Characterizing Water Pools within Reverse Micelles Using Relaxometry Techniques**

Presenting Author: Alec Beaton

Alec Beaton (Syracuse University); Alexandria Guinness (Syracuse University); John Franck (Syracuse University)

The two-component core/shell model for characterizing the water inside reverse micelles (RMs) is often difficult to verify experimentally and its details contested across different techniques. To address this dilemma, we invoke a two-pronged magnetic resonance approach: Deuterium relaxometry for probing the average hydrogen bonding and rotational motion of water, and Overhauser Dynamic Nuclear Polarization (ODNP) for probing the translational motion. Using deuterium NMR, we monitor changes in the water environment in different RM systems, including those containing guest osmolytes. We also report the first comprehensive liquid-state ODNP study on water inside RMs. Through careful selection of spin probes, we anticipate the ability to observe core and shell signal separately to develop a more complete picture of the nanoscale water dynamics.

11:50-12:10 **Integrated Platform for Production and Detection of 13C-Hyperpolarised Metabolites at Microscale**

Presenting Author: Sylwia J Barker

Sylwia Barker (University of Southampton); Laurynas Dagys (University of Southampton); Manvendra Sharma (University of Southampton); James Eills (Institute for Bioengineering of Catalonia); Malcolm Levitt (University of Southampton); Marcel Utz (University of Southampton)

In this work we demonstrate, the integrated generation and detection of a carbon-13 hyperpolarised metabolite in continuous flow in a microfluidic chip by parahydrogen induced polarisation (PHIP). The metabolite [1-13C]fumarate is produced in a nuclear hyperpolarised form by (i) introducing para-enriched hydrogen into the solution by diffusion through a semi-permeable PDMS membrane, (ii) reaction with a substrate in the presence of a ruthenium-based catalyst, and (iii) conversion of the singlet-polarised reaction product into a carbon magnetisation by the S2hM pulse sequence, achieving 13C enhancement factors of 2000; all on a single compact platform. This can be exploited for hyperpolarised multidimensional NMR experiments, which require superposition of many transients that must maintain a high level of consistency.

LUNCH-ON-YOUR-OWN
OR
ULTRA-HIGH FIELD LUNCH WORKSHOP hosted by Bruker, RSVP required, sign-up HERE
12:35-14:00

POSTER SESSION
14:00-15:45
See Poster Listings at end of document

TUESDAY, APRIL 26, 2022 - continued

TOE: Small Molecules Meet Big Challenges 1 (parallel session)
16:00-17:50, Grand DE
David Rovnyak presiding

16:00-16:25 **Small Molecule Drug Discovery Accelerated by Protein NMR Spectral Fingerprint Analysis**

Presenting Author: Andrew Namanja

Andrew Namanja (AbbVie)

Rapid assessment of target ligandability and elimination of false positives is critical in the early stages of small molecule drug discovery programs. Protein-detected 2D NMR methods are the gold standard for robust hit validation and can also find fragments that can inform on the probability of success in identifying tractable hits and binding pockets. While single point hit confirmation can be robust, the advancing of structure activity relationships (SAR) by NMR of weak binding fragments to potent leads can be a daunting and resource-intensive task. I will discuss a protein-detected 2D NMR workflow that involves single concentration affinity ranking of ligands coupled with quantitative affinity determination for fragment screening and for driving SAR using rapid NMR fingerprint analysis methods.

16:25-16:45 **Parallel NMR Supersequences: Ten 2D Spectra in a Single Measurement**

Presenting Author: Eriks Kupce

Eriks Kupce (Bruker UK Ltd); Jonathan R. J. Yong (Department of Chemistry, University of Oxford); Göran Widmalm (Department of Organic Chemistry, Arrhenius Laboratory, Stockholm University); Tim D. W. Claridge (Department of Chemistry, University of Oxford)

The principles employed in parallel NMR and MRI are applied to NMR of organic molecules. This led to design of parallel NMR supersequences yielding as many as ten 2D NMR spectra in a single measurement. Such parallel supersequences are entangled by time-sharing pulse schemes, e.g. IPAP-seHSQC, HSQCCOSY, HSQC-TOCSY and similar that are combined with modified (sequential and/or interleaved) conventional pulse sequences (modules), such as HMBC, TOCSY, COSY, CLIP-COSY, NOESY, and ROESY. A number of examples is presented where two parallel NOAH (NMR by Ordered Acquisition using 1H-detection) supersequences are applied in structure elucidation of large organic molecules, thus dramatically reducing experiment time, and increasing the sensitivity and information content that is obtained in a single NMR measurement..

16:45-17:05 **The Role of Human Protein DNAJA1 in Pancreatic Cancer – A Novel Therapeutic Target**

Presenting Author: Robert Powers

Robert Powers (University of Nebraska-Lincoln); Heidi E. Roth (University of Nebraska-Lincoln); Fatema Bhinderwala (University of Pittsburgh); Rodrigo Franco (University of Nebraska-Lincoln); You Zhou (University of Nebraska-Lincoln)

Pancreatic cancer has a dismal 5-year survival rate and is the third leading cause of cancer deaths. The cochaperone protein DNAJA1 is downregulated four-fold in pancreatic cancer cells, but little is known about its role in cancer. The impact of DNAJA1 overexpression on pancreatic ductal adenocarcinoma (PDAC) cells was evaluated using a combination of metabolomics, microscopy, and cell-based assays. We also determined the structure of the DNAJA1 J-domain and identified 11 potential protein partners and nm-□M small-molecule binders that disrupt these protein interactions. Our findings suggest a proto-oncogenic role for DNAJA1 in PDAC progression and suggest DNAJA1 may function synergistically with other proteins with altered activities in pancreatic cancer cells. DNAJA1 is a potential therapeutic target for treating pancreatic cancer.

17:05-17:25 **High Resolution NMR Measures Intact Glycan Structure and Composition of Therapeutic Monoclonal Antibodies at 13C Natural Abundance**

Presenting Author: Kang Chen

You Zhuo (US Food and Drug Administration); David Keire (US Food and Drug Administration); Kang Chen (US Food and Drug Administration)

The glycosylation on monoclonal antibodies (mAbs) is critical for drug function and safety. The covalent linkage of oligo-saccharide to a large mAb protein limited NMR data collection because of short T2. Recently, the urea denaturing approach was developed to increase glycan dynamics without cleaving off oligo-saccharide moieties. Here, the NMR method was further developed with higher resolution in the anomeric region of 2D 1H-13C HSQC spectra, which allowed the accurate assignment and quantification of clinically relevant minor glycan species containing high-mannose, afucosyl, and 1-3/1-6 branch specific galactosylation in adalimumab and trastuzumab. Therefore, the new NMR method, which is an orthogonal approach to classical glycan cleavage and mapping method, offers new potential for NMR application in therapeutic glycan research and development.

TUESDAY, APRIL 26, 2022 - continued

17:25-17:50 **Following bacterial cell metabolism with NMR spectroscopy**

Presenting Author: Guy Lippens

Guy Lippens (CNRS); Neil Cox (CNRS); Pierre Millard (INRAE); Pauline Rouan (TBI); Cyril Charlier (CNRS); Davy Sinnaeve (CNRS)

Quantitative information on the carbon isotope incorporation of metabolites is essential for flux analysis. Whereas this information is in principle present in proton NMR spectra through both direct and long-range heteronuclear coupling constants, spectral overlap and homonuclear coupling constants both hinder its extraction. We will demonstrate how pure shift 2D J-resolved NMR spectroscopy can simultaneously remove the homonuclear couplings and separate the chemical shift information from the heteronuclear coupling patterns1. We demonstrate the power of this method on cell lysates from different bacterial cultures and investigate in detail the branched chain amino acid biosynthesis.

TOF: Materials 2: Organic /Inorganic Interfaces (parallel session)

16:00-17:50, Grand F

Gaël De Paëpe presiding

16:00-16:25 **New Potentialities of NMR and DNP for the Study of Biomaterials: Experiments and Theory.**

Presenting Author: Christian Bonhomme

Christian Bonhomme (Sorbone University)

Natural biomaterials usually exhibit a high level of structural complexity. This is inherent to their hybrid nature combining organic and inorganic components at different levels (from nm to cm). In the first part of this contribution NMR and DNP crystallography concepts will be developed in the framework of pathological calcifications and their synthetic analogs. Solid state NMR/NMR methodology gives original insight on both structure and local dynamics. Most importantly, interesting phase transformations can be monitored in situ.

16:25-16:45 **Natural Abundance Satellite Transition 33S and 17O Solid State NMR by Progressive Saturation of the Proton Reservoir (PROSPR)**

Presenting Author: Tamar Wolf

Tamar Wolf (Weizmann Institute of Science); Michael John Jaroszewicz (Weizmann Institute of Science); Lucio Frydman (Weizmann Institute of Science)

Solid-state NMR techniques that reveal the chemistry of sulfur and oxygen are of broad interest, due to their widespread appearance in organic and inorganic compounds, and the key role that they play in chemical and biochemical reactions. Nonetheless, solid-state NMR studies of these species typically cannot be executed without isotope enrichment, due to low natural abundance (0.75%-0.038%), and low absolute sensitivity (≈10-5). Herein, we present the application of PRogressive Saturation of Proton Reservoir (PROSPR), a novel experiment to measure wideline powder patterns by relying on the orders-of-magnitude more sensitive 1H resonance. With these hundreds-fold sensitivity enhancements, the broad static satellite transitions patterns of 17O and 33S in ammonium sulfate at natural abundance were detected in half an hour.

16:45-17:05 **Moisture-induced CO2 species in amine-based solid adsorbents: molecular-level study from solid-state NMR and molecular modeling**

Presenting Author: Mariana Sardo

Mariana Sardo (CICECO - University of Aveiro); Rui Afonso (CICECO - University of Aveiro); Moisés L. Pinto (IST - University of Lisbon); José R. B. Gomes (CICECO - University of Aveiro); Luís Mafra (CICECO - University of Aveiro)

In recent years, our group has focused on understanding CO2 chemisorption processes in amine-modified mesoporous silicas (SBA-15) combining solid-state NMR and computer modeling. We've shown, for instance, that control over amine surface density enabled the detection of proton-transfer, among distinct CO2 species, using NMR CSA.

Here we present a comprehensive study of the influence of moisture and CO2 partial pressures on CO2 speciation upon CO2 chemisorption on SBA-15. Solid-state NMR and molecular modeling are used in tandem to shed light on the nature of such surface gas species, under dry and wet conditions, providing molecular-level details on the formation mechanism of moisture-induced CO2 species. This work extends the current understanding on the nature of chemisorbed CO2 structures under wet conditions.

TUESDAY, APRIL 26, 2022 - continued

17:05-17:25 **Tailoring Electron-electron Coupling Network for DNP at High Magnetic Field and Fast MAS**

Presenting Author: Asif Equbal

Asif Equbal (Postdoc); Songi Han (Professor)

Improving DNP efficiency at higher magnetic-fields and faster spinning is important to realize the full potential of DNP. In agreement with experimental observations, we describe the role of electron-electron coupling and, in particular, the importance of balancing dipolar and exchange couplings for efficient DNP. We also describe the (hidden) role of Thermal-Mixing DNP in high concentrations regime (> 25 mM), where intermolecular electron-electron coupling becomes significant. This is supported by strong EPR-based experimental evidence for the presence of Dipolar-Order magnetization of electron spins. Recently, BDPA and Trityl molecules were reported to exhibit pronounced high-field Thermal-Mixing DNP at concentrations between 15 mM and 80 mM. The effects of the electron-spin coupling network on nuclear spin diffusion are also discussed.

17:25-17:50 **Biomolecular Binding at Calcium Phosphate Surfaces and New NMR Crystallography Methods**

Presenting Author: Mattias Eden

Renny Mathew (Stockholm University); Baltzar Stevensson (Stockholm University); Debashis Majhi (Stockholm University); Mattias Eden (Stockholm University)

Intimate interactions between amorphous calcium phosphate (ACP) and either L-serine (Ser) or O-phospho-L-Serine (Pser) gives bone-adhesive properties of calcium phosphate cements for biomedical applications. Advanced MAS NMR experimentation was used for probing both the organic and inorganic components, as well as their interface, along with results from metadynamics simulations of a Pser/Ser molecule in water that interface with a disordered calcium phosphate surface. We will discuss the binding modes of the surface-bound molecules.

We also refined the structures of Pser and its Ca salt by DFT calculations. The structure models were validated by a recently developed NMR crystallography method that utilizes homonuclear or/and heteronuclear distances. We will also discuss applications to 1H-1H distance estimates in pharmaceuptical compounds.

Vendor Hospitality Suites
From 18:30

WEDNESDAY, APRIL 27, 2022

EARLY AM: SPIN DYNAMICS (plenary session)
07:00-08:00

07:00-08:00 **Simulation Tutorial** Part 3 of 5: **STATIC SOLIDS**

Presenter: Ilya Kuprov

WOA: Materials 3: Materials Meet Big Challenges (parallel session)
08:30-10:20, Grand DE
Danielle Laurencin presiding

08:30-08:55 **Ex Situ and in Situ Nuclear Magnetic Resonance of Materials for Energy Storage**

Presenting Author: Elodie Salager

Ludivine Afonso de Araujo (Technocentre Renault & CNRS-CEMHTI); Ghenima Oukali (CNRS-CEMHTI); Charles-Emmanuel Dutoit (CNRS-CEMHTI); Benjamin Porcheron (CNRS-CEMHTI); David Sicsic (Technocentre Renault); Encarnacion Raymundo Piñero (CNRS-CEMHTI); Vincent Sarou-Kanian (CNRS-CEMHTI); Michael Deschamps (CNRS-CEMHTI); Elodie Salager (CNRS-CEMHTI)

Energy storage is at the heart of the necessary transition towards greener energies. Understanding the limiting factors of electrochemical devices is essential to develop next generation solution for electrochemical storage.

Ex situ NMR studies of solid electrodes and electrolytes of rechargeable batteries provide structural and/or dynamical insight. In situ NMR was also recorded in full supercapacitors to catch the influence of various nanoporous electrodes on the behaviour of the ions in the electrolyte upon charge and discharge. We will finally report in situ NMR of Li-ion battery devices containing commercial electrodes at low temperature and high charge. Experimental development and a careful analysis of the data helped rationalizing the early appearance of plated lithium, an ageing factor in commercial batteries.

08:55-09:15 **Elucidating Polysaccharide-Aromatic Interactions in Wood Cell Walls by Solid-State NMR**

Presenting Author: Wancheng Zhao

Wancheng Zhao (Louisiana State University); Alex Kirui (Louisiana State University); Fabien Deligey (Louisiana State University); Hui Yang (Pennsylvania State University); Xue Kang (Louisiana State University); Frederic Mentink-Vigier (National High Magnetic Field Laboratory, Florida State University); Tuo Wang (Louisiana State University)

Plant cell walls, as an inexhaustible resource of biomaterials and biofuel, include many carbohydrates and an aromatic polymer named lignin. These major components of lignocellulosic biomass are substantially underrepresented in high-resolution solid-state structural studies. Here, we elucidate the nanoscale assembly of lignocellulosic components in wood stems (Eucalyptus, Poplar, and Spruce) using solid-state NMR and dynamic nuclear polarization (DNP) approaches. Our results have shown that cellulose-lignin contacts are absent in grass but abundant in wood. Woody plants have larger cellulose bundles and more flat-ribbon xylan. Softwood has unique cell wall architecture, with all molecules homogeneously mixed on the nanoscale. These structural principles will guide the rational design of more digestible plants and more efficient biomass-conversion pathways.

WEDNESDAY, APRIL 27, 2022 - continued

09:15-09:35 **Hyperpolarized Solution-State NMR Spectroscopy with Optically Polarized Crystals**

Presenting Author: Anna J. Parker

Tim R. Eichhorn (NVision Imaging Technologies); Anna Parker (NVision Imaging Technologies); Felix Josten (NVision Imaging Technologies); Christoph Mueller (NVision Imaging Technologies); Jochen Scheuer (NVision Imaging Technologies); Jakob M. Steiner (NVision Imaging Technologies, Paul Scherrer Institut); Martin Gierse (NVision Imaging Technologies, University of Ulm); Jonas Handwerker (NVision Imaging Technologies); Michael Keim (NVision Imaging Technologies); Sebastian Lucas (NVision Imaging Technologies); Mohammad Usman Qureshi (NVision Imaging Technologies); Alastair Marshall (NVision Imaging Technologies); Alon Salhov (NVision Imaging Technologies, Hebrew University); Yifan Quan (MIT); Pinelopi Moutzouri (École Polytechnique Fédérale de Lausanne); Federico De Biasi (École Polytechnique Féderale de Lausanne); Jan Binder (NVision Imaging Technologies); Kay D. Jahnke (NVision Imaging Technologies); Philipp Neumann (NVision Imaging Technologies); Stephan Knecht (NVision Imaging Technologies); John W. Blanchard (NVision Imaging Technologies); Martin B. Plenio (Ulm University); Fedor Jelezko (Ulm University); Lyndon Emsley (École Polytechnique Fédérale de Lausanne); Christophoros C. Vassiliou (NVision Imaging Technologies); Patrick Hautle (Paul Scherrer Institute); Ilai Schwartz (NVision Imaging Technologies)

Nuclear spin hyperpolarization provides a promising route to overcome the challenges imposed by the limited sensitivity of NMR. Here we demonstrate that dissolution of spin-polarized pentacene-doped naphthalene crystals enables transfer of polarization to target molecules at room temperature and moderate to high magnetic fields (1.45 T - 9.4 T). This polarization source is chosen for two properties: solubility in common organic solvents and 1H relaxation times T1 sufficiently long for storage, enabling the decoupling of the source polarization process from the transfer of source polarization to target nuclei. We observe NMR signals enhanced by factors between -200 and -1730 (1.45 T) for a range of small molecules and enhancements of >10 on target 1H sites of diethyl [difluoro(trimethylsilyl)methyl]phosphonate (400 MHz).

09:35-09:55 **Quantum sensing with optically hyperpolarized nuclei**

Presenting Author: Ashok Ajoy

Ashok Ajoy (UC Berkeley)

“Quantum sensor” technologies have opened attractive new applications stemming from the sensitive detection of magnetic fields. This is typified by the Nitrogen Vacancy (NV) center in diamond, that has allowed the nanoscale sensing of spins in materials and molecules through optical means. I will describe a complementary modality of how such quantum sensors could assist in the injection of spin polarization into surrounding nuclei, “hyperpolarizing” them into athermal spin states that are far from equilibrium. I will show that this portends the ability to construct new sensor platforms from hyperpolarized nuclear spins, exploiting their remarkable spin coherence properties at room temperature. This could ultimately yield interesting new “deployable” magnetic resonance sensors with high chemical resolving power and submicron spatial resolution.

09:55-10:20 **"NMR Crystallization": New In-situ NMR Techniques for Time-resolved Monitoring of Crystallization Processes**

Presenting Author: Kenneth D. M. Harris

Kenneth Harris (Cardiff University)

Motivated by exploring the time-evolution of crystallization processes and understanding fundamental aspects of crystallization systems, we are developing in-situ solid-state NMR strategies to monitor the structural evolution of the solid phase in crystallization systems, including the identification of transitory amorphous phases and intermediate crystalline phases (polymorphs) on the pathway towards the final crystallization product. In addition, we have developed a strategy (called "CLASSIC NMR") to study the complementary changes that occur in both the solid phase and the liquid phase as a function of time during crystallization from solution. The lecture will give an overview of the application of these NMR techniques to monitor crystallization processes, highlighting examples of crystallization of organic materials, metal-organic frameworks and mineral phases.

WEDNESDAY, APRIL 27, 2022 - continued

WOB: Theory and Computation 2: Modern Computation & Biomolecular NMR (parallel session)

08:30-10:20, Grand F
Leonard Mueller presiding

08:30-08:55 **NMR of complex native assemblies**

Presenting Author: Francesca M Marassi

Francesca Marassi (Sanford Burnham Prebys Institute)

Structural biology can provide fundamental information for understanding how biomolecules function, but the samples are often highly simplified versions of the complex native environment. We will describe experimental and computational approaches for studying biomolecules in native environments. Virulence phenotypes can be expressed in model bacterial models, and isolated cell envelopes expressing 15N and 13C labeled protein yield atomic-resolution NMR spectra that allow us to probe structure and protein-protein interactions in situ. The data shed light on the interactions of bacteria with human serum and provide a platform for advancing structure-activity NMR studies in the native environment, including its various physiological modulations that can be elicited by factors such as temperature, antibiotics, or exposure to serum.

08:55-09:15 **Data, Meet Computation**

Presenting Author: Jeffrey C. Hoch

Jeffrey Hoch (UConn Health); Kumaran Baskaran (UConn Health); D Levi Craft (UConn Health); Hamid Eghbalnia (UConn Health); Michael R. Gryk (UConn Health); Mark W. Maciejewski (UConn Health); Adam D. Schuyler (UConn Health); Jonathan R. Wedell (UConn Health); Colin W. Wilburn (UConn Health)

BMRB is the open, international repository for NMR data on biomolecules. NMRbox is a computational platform for NMR data processing and analysis with 200+ software packages across all areas of NMR and related fields. We have implemented a data lake on the NMRbox platform that provides local snapshots of the BMRB archive, as well as snapshots of the PDB, NCBI non-redundant sequence, and AlphaFold archives. Combining these data resources with the computational resources of NMRbox simplifies and accelerates studies requiring federation of the separate archives, for example machine learning approaches that are poised to have transformative impact.

09:15-09:35 **The making of DEERNet: why and how do neural networks work in magnetic resonance data processing?**

Presenting Author: Ilya Kuprov

Jake Keeley (University of Southampton); Tajwar Choudhury (University of Southampton); Laura Galazzo (University of Geneva); Enrica Bordignon (University of Geneva); Akiva Feintuch (Weizmann Institute); Daniella Goldfarb (Weizmann Institute); Hannah Russell (University of St Andrews); Michael J. Taylor (University of St Andrews); Janett E. Lovett (University of St Andrews); Andrea Eggeling (ETH Zurich); Luis Fabregas Ibanez (ETH Zurich); Katharina Keller (ETH Zurich); Maxim Yulikov (ETH Zurich); Gunnar Jeschke (ETH Zurich); Ilya Kuprov (University of Southampton)

We have recently released a neural network based data processing package for pulsed dipolar spectroscopy (DEER and RIDME sequences) that matches or exceeds the performance of state-of-the-art deterministic and statistical processing methods. The level of performance (a few hours of unattended training) relative to the existing tools (a few years of human developer effort) was unexpected; this prompted a deeper investigation.

After finding a mathematical procedure that clarifies internal mechanics of fully connected neural nets, we found considerable internal sophistication in DEERNet: the network spontaneously invents a bandpass filter, a notch filter, a frequency axis rescaling transformation, frequency division multiplexing, group embedding, spectral filtering regularisation, and a map from harmonic functions into Chebyshev polynomials. This is fascinating and slightly scary.

09:35-09:55 **Structural Fingerprinting of Short Oligonucleotide Therapeutics by Solution NMR Spectroscopy**

Presenting Author: Owen B Becette

Owen Becette (Institute for Bioscience and Biotechnology Research); John P. Marino (NIST/IBBR); Robert G. Brinson (NIST/IBBR)

Short oligonucleotide therapeutics are an emerging class of biopharmaceuticals to treat diseases that are inaccessible to traditional small molecule- and protein-based approaches. Despite the growing interest in short oligonucleotide therapeutics, there are a lack of high-resolution techniques needed to rigorously characterize these molecules, to establish metrics for structure-related quality attributes during drug development. Here we present high-resolution NMR spectroscopy experiments to structurally “fingerprint” short oligonucleotides at natural isotopic abundance and typical formulation conditions. In addition to standard 1H, 13C, and 15N correlations, short oligonucleotides contain 19F and 31P which provide additional NMR handles for structural probing. We demonstrate the application of NMR methods to structurally “fingerprint” short oligonucleotide therapeutics using model antisense oligonucleotides (ASO) and short interfering ribonucleic acids (siRNAs).

WEDNESDAY, APRIL 27, 2022 - continued

09:55-10:20 **Faster prediction of solid-state NMR chemical shifts without compromising accuracy**

Presenting Author: Gregory Beran

Gregory Beran (University of California Riverside)

This talk will focus on two computationally inexpensive strategies for improving chemical shift prediction accuracy. The first approach demonstrates how existing GIPAW calculations can be improved via a fast, straightforward gas-phase correction to the chemical shifts. The second approach employs so-called Δ-machine learning to achieve DFT-quality chemical shift predictions 1-2 orders of magnitude faster than DFT. Although these Δ-ML models are somewhat slower than pure ML models, they can achieve considerably better accuracy. We demonstrate that the accuracy of our Δ-ML approach is on par with DFT, in the sense that the Δ-ML errors in reproducing the DFT shieldings it was trained against are considerably smaller than those typically found when comparing DFT chemical shifts against experiment.

COFFEE BREAK with Exhibit Booths
10:10-10:45

WOC: Biomolecular Solutions 2: Sensors and Interactions (parallel session)
10:45-12:35, Grand DE
Jeffrey Peng presiding

10:45-11:10 **In-cell NMR: a powerful approach for studying protein folding and for drug screening**

Presenting Author: Lucia Banci

Lucia Banci (University of Florence)

In-cell NMR represents one of the highest impact applications of magnetic resonance.

These experiments allow to obtain information on the conformational and functional properties of biomolecules at atomic resolution in conditions as close as possible to the physiological ones. In-cell NMR also allows to monitor protein-protein interactions and to follow functional processes in real time as well as to perform drug screening at cellular level.

Methodological aspects and innovations will be discussed and a few examples presented. Particular focus will be on the meaningful differences observed on biological molecules in living cells versus those in vitro, with respect to the oxidation and folding states, and to the cellular uptake and target binding in the cellular milieu by drugs and leads.

11:10-11:30 **Probing molecular details of cosolute-protein interactions using paramagnetic relaxation enancement**

Presenting Author: Yusuke Okuno

Yusuke Okuno (National Institute of Health); Charles D. Schwieters (National Institute of Health); G. Marius Clore (National Institute of Health)

The addition of small molecules (cosolutes) in solution can perturb protein stability, structure, and biological activity. Yet, the molecular mechanism whereby cosolutes denature/stabilize proteins is still a subject of considerable debate. We developed a new NMR experimental technique to probe cosolute-protein interactions based on paramagnetic relaxation enhancement. We applied this technique to the model protein drkN SH3 to study how cosolutes interact on the surface of folded and unfolded states. Two denaturing cosolutes with different electrostatic charges are employed and showed that both cosolutes interact mostly on unfolded states while the interactions are significant only in the loop regions on the folded state. The experimentally derived effective near-surface potential (ENS) are also calculated for both folded and unfolded states.

WEDNESDAY, APRIL 27, 2022 - continued

11:30-11:50 **Global structural dynamics as sensors of molecular events in non-ribosomal peptide synthetases**

Presenting Author: Subrata H. Mishra

Subrata Mishra (Johns Hopkins University School of Medicine); Aswani Kancherla (Johns Hopkins University School of Medicine); Kenneth Marincin (Johns Hopkins University School of Medicine); Guillaume Bouvignies (ENS - Laboratoire des BioMolécules); Santrupti Nerli (University of California, Santa Cruz); Nikolaos Sgourakis (University of Pennsylvania); Daniel Dowling (University of Massachusetts Boston); Dominique Frueh (Johns Hopkins University School of Medicine)

What role does intra-domain protein dynamics play in multi-domain protein communication? Our investigations of nonribosomal peptide synthetase (NRPS) reveal that intra-domain dynamics drives inter-domain interactions. NRPSs are multidomain enzymatic systems that synthesize complex natural products from simple substrates via transient sequential interactions between domains. How these sequential interactions are orchestrated during synthesis has been cryptic as interpretations have primarily relied on static structural snapshots.

Using solution NMR, we demonstrate that global structural dynamics drives substrate-dependent communication and allosteric responses. Impeding the intra-domain dynamics severs the allosteric response and molecular recognition. Our results establish global structural fluctuations as sensors of molecular events that can remodel inter-domain interactions, and they provide new perspectives on mechanisms of allostery, protein communication, and NRPS synthesis.

11:50-12:10 **Role of conformational entropy in molecular evolution of bacterial transcriptional repressors**

Presenting Author: Giuliano T. Antelo

Giuliano Antelo (Fundación Instituto Leloir); Cristian Pis-Diez (Fundación Instituto Leloir); Hongwei Wu (Chemistry, Indiana University); Matías Villarruel-Dujovne (Fundación Instituto Leloir); David P. Giedroc (Chemistry, Indiana University); Daiana A. Capdevila (Fundación Instituto Leloir)

Protein dynamics, unveiled by NMR spectroscopy, have risen as a key factor in the modulation of protein function. In a member of the ArsR (arsenic repressor) family of bacterial transcription factors, key players in the resistance mechanisms of pathogens to antibiotics and host-derived stress, we have previously shown that the allosteric connection between the DNA-binding site and the stress-recognition site relies not in global structural changes, but in a redistribution of fast internal motions inside the protein. Here, we generalize this principle, and we present our results in a novel sulfide-sensor in which we have collected numerous crystal structures and 3D-NMR spectra that allowed for the calculation of the methyl order parameter (O2axis) and its conformational entropy in several states.

12:10-12:35 **Biomolecular Electrostatics by NMR**

Presenting Author: Junji Iwahara

Binhan Yu (University of Texas Medical Branch); Channing C. Pletka (University of Texas Medical Branch); Junji Iwahara (University of Texas Medical Branch)

Biomolecular electrostatics are important for our understanding of molecular functions of proteins and nucleic acids. Accurate information about electrostatic interactions is also crucial for success in drug development and protein engineering. However, electrostatic interactions involving mobile charges are difficult to investigate. Recently, we developed some NMR methods for investigating biomolecular electrostatics. This presentation will cover two methods we recently published. One is a paramagnetic NMR-based method for de novo determination of near-surface electrostatic potentials around proteins and nucleic acids. The other is a diffusion NMR spectroscopy-based method for investigating counterions around biomolecules.

WOD: In-Vivo / Ex-Vivo 3 (parallel session)
10:45-12:35, Grand F
Matthew Rosen presiding

10:45-11:10 **Chemical Exchange Saturation Transfer (CEST) MRI: from in-vivo imaging to pathology assessment.**

Presenting Author: Elena Vinogradov

Elena Vinogradov (University of Texas Southwestern Medical Center)

Chemical Exchange Saturation Transfer (CEST) employs selective saturation of the exchanging protons and subsequent detection of the water signal decrease to create images that are weighted by the presence of metabolites or pH. Recently, it has been gaining recognition for its ability to detect metabolic alterations in a variety of diseases from cancer to neurodegeneration. Here, we will describe aspects of translating CEST to reliable clinical applications at 3Tesla and discuss its potential uses in human oncology, specifically breast and kidney cancers, as well as is in neurodegeneration. Next, we will discuss correlative studies between the in-vivo CEST imaging and ex-vivo tissue analysis (e.g. histopathology), aimed at elucidating origins of the observed CEST contrast alterations.

WEDNESDAY, APRIL 27, 2022 - continued

11:10-11:30 **Radio Frequency Sweeps at uT Fields for Parahydrogen Induced Polarization of Biomolecules**

Presenting Author: Alastair Marshall

Alastair Marshall (NVision Imaging Technologies); Alon Salhov (NVision Imaging Technologies GmbH); Christoph Müller (NVision Imaging Technologies GmbH); Martin Gierse (NVision Imaging Technologies GmbH); Anna J. Parker (NVision Imaging Technologies GmbH); Jochen Scheuer (NVision Imaging Technologies GmbH); Michael Keim (NVision Imaging Technologies GmbH); Sebastian Lucas (NVision Imaging Technologies GmbH); Christophoros Vassilou (NVision Imaging Technologies GmbH); John Blanchard (NVision Imaging Technologies GmbH); Philipp Neumann (NVision Imaging Technologies GmbH); Fedor Jelezko (NVision Imaging Technologies GmbH); Alex Retzker (NVision Imaging Technologies GmbH); Ilai Schwartz (NVision Imaging Technologies GmbH); Stephan Knecht (NVision Imaging Technologies GmbH)

Magnetic resonance imaging of the metabolic processes within the human body opens up new methods of studyingcancers. It is accomplished through the monitoring of the 13C NMR signal of a metabolite, which has an inherently low signal strength. Hyperpolarization, where parahydrogen is chemically added to an unsaturated precursor molecule, generating an entangled spin state in the product molecule, offers a solution. The spin-order of the hyperpolarized protons is converted into observable 13C magnetization using magnetic field sweeps. However, when using deuterated precursor molecules, the efficiency is severely reduced. We demonstrate that using a radio-frequency sweep, a 50% stronger signal can be measured compared to the protonated molecule. We demonstrate the technique on a number of molecules.

11:30-11:50 **HX- enabled fast scanning for simultaneous NMR detection of H2O2 and organic hydroperoxides**

Presenting Author: Tayeb Kakeshpour

Tayeb Kakeshpour (NIH/NIDDK); Belhu Metaferia (NIH/NIDDK); Richard N. Zare (stanford); Adriaan Bax (NIH/NIDDK)

Despite the importance of H2O2 in various fields, methods typically used for its quantification are indirect (chemical) which can suffer from reaction sluggishness or the presence of interfering species. We demonstrate the use of NMR for direct quantification of H2O2 down to the mid-nanomolar range. Our method takes advantage of the fast hydrogen exchange (HX) between water and H2O2 molecules for eliminating long inter-scan delays, permitting fast scanning for S/N enhancement. We demonstrate the method for H2O2 quantification in air, rain, exhaled breath condensate, urine, saliva, and blood. We also show that various organic hydroperoxides can be simultaneously quantified in a mixture.

11:50-12:10 **Comprehensive Multiphase NMR: a powerful and versatile tool to observe molecular processes**

Presenting Author: Rajshree Ghosh Biswas

Rajshree Ghosh Biswas (University of Toronto); Ronald Soong (University of Toronto); Mark Croxall (University of Toronto); Reece Lawrence (University of Toronto); Paris Ning (University of Toronto); Daniel Schmidig (Bruker BioSpin AG); Peter De Castro (Bruker BioSpin AG); Stephan Graf (Bruker BioSpin AG); Sebastian Wegner (Bruker Biospin GmbH); Falko Busse (Bruker Biospin GmbH); Cynthia Goh (University of Toronto); Myrna Simpson (University of Toronto); Andre Simpson (University of Toronto)

Most natural systems are heterogenous mixtures consisting of all phases (solids, liquids, gels) and requires a non-invasive technique capable of observing molecular interactions in situ to understand the entire system. Observing all phases using traditional NMR approaches often overlook or even destroys (i.e., grinding and dry packing) interactions between individual phases, transformation of one phase to another, and movement of compounds between phases. Here, Comprehensive Multiphase (CMP-NMR) is used to investigate a variety of complex multiphase processes to obtain information about the sample in its entirety. This study will explore processes such as the photocatalytic degradation of phenol, biofuel composition and extraction efficiency under varying conditions, and metabolic changes in Daphnia magna during aging and reproduction from a multiphase perspective.

12:10-12:35 **Current efforts to push the limits of short-T2 imaging in humans**

Presenting Author: Klaas Pruessmann (ETH Zurich)

LUNCH-ON-YOUR-OWN
12:35-14:00

POSTER SESSION
14:00-15:45
See Poster Listings at end of document

WOE: Tutorial Session: Biomolecular NMR, 17O NMR, and Building a Resonant Circuit
16:00-18:00, Grand DE

16:00-16:40 **Intrinsically Disordered Proteins by NMR Spectroscopy**
Presenting Author: Isabella Felli (University of Florence)

16:40-17:20 **Oxygen-17 NMR spectroscopy: Examples of Strategies for Achieving Higher Sensitivity**
Presenting Author: Danielle Laurencin (CNRS)

17:20-18:00 **Practical guide to building and safety testing RF coils**
Presenting Author: Jason Stockmann (Massachusetts General Hospital)

Vendor Hospitality Suites
From 18:30

THURSDAY, APRIL 28, 2022

EARLY AM: SPIN DYNAMICS (plenary session)
07:00-08:00

07:00-08:00 **Simulation Tutorial** Part 4 of 5: **SPINNING SOLIDS**

Presenter: Ilya Kuprov

ThOA: Biomolecular Solutions 3: Structure and Dynamics (parallel session)
08:30-10:20, Grand DE
Rieko Ishima presiding

08:30-08:55 **Gating, Selectivity and Allostery in the NaK Channel**

Presenting Author: Katherine Henzler-Wildman

Vilius Kurauskas (UW-Madison); Adam Lewis (UW-Madison); Marco Tonelli (UW-Madison); Katherine Henzler-Wildman (UW-Madison)

Correlation of structural and functional states is crucial for understanding how ion channels selectively conduct ions in response to stimuli. High resolution crystal structures of NaK, a non-selective cation channel, bound to Na+ and K+ suggest a rigid selectivity filter (SF) structure, while comparison full-length NaK (FL-NaK) and a truncated form (NaKΔ18) suggest a simple model of steric gating at the helix bundle crossing (HBC). These models are not fully consistent with MD simulations, solid-state NMR data or channel flux measurements. Here, we use solution NMR experiments to characterize how bound Na+ vs. K+ affects the NaK SF structure and dynamics and assess the HBC model for NaK gating for NaK solubilized in lipid bicelles.

08:55-09:15 **Methodological advances for multi-site exchange in Cadherin-11**

Presenting Author: Hans Koss

Hans Koss (Columbia University); Barry H. Honig (Columbia University); Lawrence S. Shapiro (Columbia University); Arthur G. Palmer (Columbia University)

Dimerization of Cadherin-11, and probably more generally of type II Cadherins, involves coupled unfolding and strand-swapping. Our recent comprehensive analysis of Cadherin-11-EC1 based on various relaxation dispersion (RD) experiments has delivered a quantitative description of a three-site kinetic connectivity. The involved conformations, varying in the degree of A-strand exposure, also are coupled to additional conformational states on very fast and very slow timescales. The RD data analysis has applied some of our previous theoretical findings. Herein, we report on experimental advances to characterize very fast and very slow chemical exchange. As an example, we have used a simple pulse sequence to efficiently record exchange-suppressed Hahn-Echo relaxation, characterizing the previously identified very fast exchange process at different concentrations and temperatures.

09:15-09:35 **Increased Protein Dynamics Defines Ligandability**

Presenting Author: Lukasz Jaremko

Vladlena Kharchenko (KAUST); Brian Linhares (UMICH); Megan Borregard (UMICH); Iwona Czaban (KAUST); Jolanta Grembecka (UMICH); Mariusz Jaremko (KAUST); Tomasz Cierpicki (UMICH); Lukasz Jaremko (KAUST)

We demonstrate that analysis of protein dynamics using NMR, largely neglected in drug-discovery fields, can both detect and localize a highly ligandable binding site in new protein targets. We evaluated the ligandability of proteins in the BTB-domain family, which mediate protein-protein interactions (PPIs). BTB domain-containing proteins are abundant in humans and are involved in gene regulation. BTBs are attractive targets for drug discovery. To date, small molecule inhibitors have only been reported for the BCL6-BTB domain, begging the question of whether highly conserved paralogues can be also targeted. Here, we present an innovative approach that allows unambiguous identification of ligandable targets for subsequent drug discovery campaigns. We believe that profiling protein dynamics can also be expanded to other protein classes.

THURSDAY, APRIL 28, 2022 - continued

09:35-09:55 **Insights into the structural and functional adaptation of an adapter protein Skp1 (S-Phase Kinase associated Protein-1)**

Presenting Author: Amrita Bhattacharya

Amrita Bhattacharya (IIT Bombay)

Skp1(S-phase kinase-associated protein 1 - Homo sapiens) is an adapter protein of SCF complex, which links the constant components (Cul-1-RBX) and the variable receptor (F-box proteins) in Ubiquitin E3 ligase. The solution structure reveals that Skp1 backbone dynamics reveals that Skp1 is composed of an ordered core and disordered C-term and loops in absence of the binding partner F-box. A comprehensive comparison with previously available X-ray, Cryo-EM structures of Skp1 in the SCF complex reveals the structural changes happening upon complex formation. These changes would facilitate Skp1 to adapt to the biological function of recruiting 69-different F-box proteins, one at a time. Further CPMG and CEST-based studies reveal that the binding mode is conformational exchange.

09:55-10:20 **Structure of NPSL2, a regulatory element in the oncomiR-1 RNA**

Presenting Author: Sarah C Keane

Sarah Keane (University of Michigan); Yaping Liu (University of Michigan); Aldrex Munsayac (University of Michigan); Ian Hall (University of Michigan)

MicroRNAs are small non-coding RNAs that post-transcriptionally regulate gene expression. To maintain proper microRNA expression levels, the enzymatic processing of primary and precursor microRNA elements must be strictly controlled. However, the molecular determinants underlying this strict regulation of microRNA biogenesis are not fully understood. We are investigating the differential processing of oncomiR-1, a polycistronic primary microRNA that is enriched in many cancers. NPSL2 is an auxiliary hairpin within the oncomiR-1 transcript predicted to adopt multiple structures to regulate the processing of a downstream microRNA element. We determined the solution structure of NPSL2 and have identified an alternative base pairing conformation. We are expanding our structural studies to build a comprehensive view of the oncomiR-1 structure.

ThOB: Instrumentation, Hyperpolarization, & Eclectica 2: Modern Materials Processing (parallel session)

08:30-10:20, Grand F

Thorsten Maly presiding

08:30-08:55 **MR and MRI of Materials with a Parallel Plate RF Probe and Variable Field Superconducting Magnet**

Presenting Author: Bruce Balcom

Bruce Balcom (University of New Brunswick)

Fuel cells, batteries and other electrochemical cells have a natural thin film geometry. The parallel plate resonator (PPR) is well suited to such systems and permits a substantial increase in sample size over other RF probes.

New generations of magnets are maintained superconducting by active cooling with a cryocooler, rather than cryogens. Such magnets quench if active cooling fails through power loss. An integrated magnet power supply permits ready quench recovery, but also permits facile static field change to control susceptibility effects, or to change the nucleus under investigation, with no change in RF probe frequency.

We introduce the PPR and variable field magnet, then show the advantages of their combined use for multi nuclei studies of batteries.

08:55-09:15 **Dielectric Constants of Materials for Optimized Dynamic Nuclear Polarization from 70 to 970 GHz and Subsequent MAS-DNP NMR Probe Development**

Presenting Author: Faith Scott

Faith Scott (National High Magnetic Field Laboratory); Thierry Dubroca (National High Magnetic Field Laboratory); Frederic Mentink-Vigier (National High Magnetic Field Laboratory, Florida State University); Joanna R. Long (National High Magnetic Field Laboratory)

Dynamic Nuclear Polarization (DNP) increases NMR sensitivity via unpaired electron spins added to the sample. As the resonance frequency for a radical is ~658 times that of 1H nuclei, modern DNP spectrometers operate in the hundreds of GHz range. To design better hardware, knowledge about the dielectric properties of materials and samples used in DNP is critical. In this study we have measured the dielectric constants of many materials used in DNP rotors such as sapphire, zirconia, and SiAlON, and DNP matrices such as such as glycerol-water mixture in frequency ranges from 70 GHz to 970 GHz. These constants were applied to design new rotors with different materials. We then rationalized their experimental performance via CST microwave simulations.

THURSDAY, APRIL 28, 2022 - continued

09:15-09:35 **Diamond Rotors for DNP MAS NMR**

Presenting Author: Natalie Golota

Natalie Golota (Massachusetts Institute of Technology); Zach Fredin (Massachusetts Institute of Technology); Daniel Banks (Massachusetts Institute of Technology); Salima Bahri (Massachusetts Institute of Technology); Neil Gershenfeld (Massachusetts Institute of Technology); Robert Griffin (Massachusetts Institute of Technology)

Single crystal CVD diamond rotors can enable unprecedented advances in both the sensitivity and resolution of magic angle spinning (MAS) NMR under ambient and Dynamic Nuclear Polarization (DNP) conditions. CVD diamond has extremely high tensile and elastic moduli, is nearly transparent at THz frequencies, and has exceptional thermal conductivity. While diamond is an optimal material for DNP MAS rotors, significant fabrication challenges have prevented the realization of diamond rotors. Accordingly, we developed a laser micromachining process to successfully fabricate 0.7 mm diamond rotors. We demonstrate MAS results of up to 123 kHz using nitrogen spinning gas at room temperature without rotor damage, and stability of two separate rotors at 111 kHz with a standard deviation of <4 Hz.

09:35-09:55 **MAS Spherical Shell Rotors and Spherical Solenoid Coils Boost RF Homogeneity and NMR Sensitivity**

Presenting Author: Chukun Gao

Chukun Gao (ETH Zurich); Pin-Hui Chen (ETH Zurich, lab of Physical Chemistry); Alexander Däpp (ETH Zurich, lab of Physical Chemistry); Michael Urban (ETH Zurich, lab of Physical Chemistry); Ronny Gunzenhauser (ETH Zurich, lab of Physical Chemistry); Alexander Barnes (ETH Zurich, lab of Physical Chemistry)

MAS spherical rotors, compared to the cylindrical ones, show advantages such as easy fabrication, simple sample exchange, and most importantly, high spinning frequency with minimal risk of rotor crashes. However, the MAS spheres suffer from the poor NMR sensitivity due to the small sample volume and large RF coil. We solve this problem by machining hollowed rotor with 0.5 mm wall thickness, and using a spherical solenoid coil tightly wrapped around the rotor for spinning and signal detection. A ring stator that is external to the coil supplies spinning gas. We also show that with the same sample volume and wall thickness, the spherical rotor has similar NMR sensitivity compare to the traditional cylindrical rotor.

09:55-10:20 **Implanted Ion Beta-Detected NMR as a Probe of Materials**

Presenting Author: W.Andrew MacFarlane

W. MacFarlane (UBC)

Beta-detected NMR is an exotic form of nuclear magnetic resonance that uses the asymmetric property of radioactive beta decay, instead of the induced EMF in a pickup coil, to detect the signal. In this talk, I will briefly present the general aspects of the method and its implementation at TRIUMF and review some notable recent results in crystalline solids, thin films, soft matter and room temperature ionic liquids.

COFFEE BREAK with Exhibit Booths
10:20-10:45

THURSDAY, APRIL 28, 2022 - continued

ThOC: Small Molecules Meet Big Challenges 2: Biomedical Applications (parallel session)

10:45-12:35, Grand DE

Jeffrey Peng presiding

10:45-11:10 **Enabling the application of Fluorine NMR to study high molecular weight biological systems**

Presenting Author: Haribabu Arthanari

Andras Boeszoermenyi (Harvard Medical School); Denitsa Radeva (Faculty of Chemistry and Pharmacy, Sofia University, Sofia, Bulgaria); Abhinav Dubey (Harvard Medical School); Thibault Viennet (Harvard Medical School); Vladimir Gelev (Faculty of Chemistry and Pharmacy, Sofia University, Sofia, Bulgaria); Sandeep Chhabra (Harvard Medical School); Helena Kovacs (Bruker Biospin, Fällanden, Switzerland); Wolfgang Bermel (Bruker Biospin Ettlingen, Germany); Clemens Anklin (Bruker Biospin, Billerica, MA, USA); Gerhard Wagner (Harvard Medical School); Ilya Kuprov (School of Chemistry, University of Southampton, Highfield, Southampton, UK); Koh Takeuchi (8) Molecular Profiling Research Center for Drug Discovery, National Institute of Advanced Industrial Science and Technology, Tokyo, Japan.); Haribabu Arthanari (Harvard Medical School)

The advantages of fluorine NMR, including the sensitivity of its chemical shift to the electronic environment, high gyromagnetic ratio, NMR active at natural abundance, and the absence of fluorine in biological systems have been harnessed in drug discovery. However, its application to study biological systems has been limited, predominantly because the CSA of fluorine induces rapid relaxation in large molecular weight systems. We identified a favorable interference between two relaxation mechanisms (DD and CSA) that helps disperse the broad fluorine resonances using the frequency of carbon that is directly attached to fluorine. This 13C-19F-TROSY effect manifests in aromatic systems with applications to proteins and nucleic acids. The talk will introduce the 13C-19F-TROSY effect and highlight some of the emerging applications.

11:10-11:30 **NMR of Glycosylated Proteins: Glycan Interactions in CEACAM1-IgV**

Presenting Author: James H Prestegard

Jim Prestegard (University of Georgia)

Glycans of glycoproteins play many important functions, including regulating access to protein surfaces in cell-pathogen and cell-cell interactions. Conformations for these glycans can be generated in MD trajectories, but are they all populated? Here, we explore preferred glycan conformations for the N-terminal domain of the cell-surface signaling molecule, CEACAM1. A lanthanide-binding loop is inserted in the protein sequence to provide PCS and PRE data. Assignment of crosspeaks from 13C enriched valine and alanine methyl groups using a sparse-labeling assignment tool (ASSIGN_SLP_GUI) allows determination of a susceptibility tensor, and screening of selected glycan conformers with predicted PREs and PCSs for glycan resonances. We find that conformers having glycan N-acetyl methyls interacting with hydrophobic pockets on the protein surface are frequently sampled.

11:30-11:50 **Phosphates Form Spectroscopically Dark State Assemblies in Common Aqueous Solutions**

Presenting Author: Jiaqi Lu

Jiaqi Lu (NYU); Joshua Straub (UCSB); Mesopotamia Nowotarski (UCSB); Song-I Han (UCSB); Alexej Jerschow (NYU)

Polyphosphates play a ubiquitous role in biology, from structural components to energy storage, however the solution phase space of phosphate species appears more complex than previously known. We present evidence that phosphate species including orthophosphates, pyrophosphates and adenosine phosphates associate into dynamic assemblies in dilute solutions that are spectroscopically 'dark'. NMR indicates that a majority population of phosphates remain as unassociated ions in exchange with these, spectroscopically invisible, assemblies. The formation of these weakly associated soft assemblies appears entropically driven via phosphate dehydration, observed through diffusion ordered spectroscopy and modulated by molecular crowders and salts. These results suggest that hidden phosphate assemblies can occur under biologically relevant conditions, lending more insight into the interpretation of phosphate-containing pathways and species.

11:50-12:10 **A Pipeline for Accelerating Drug Discovery: Screening and Affinity-Ranking of Fluorinated Ligands with CSAR**

Presenting Author: Alvar Gossert

Simon Ruedisser (ETH Zurich); Gabriela Stadler (Novartis AG); Helena Kovacs (Bruker AG); Alvar Gossert (ETH Zurich)

We have established an efficient drug discovery pipeline based on the newly developed method CSAR (Chemical Shift anisotropy-based Affinity Ranking). It consists of a CSAR-library of fluorinated fragments, which can be screened in a day. Subsequently, CSAR allows comparing affinities of ligands with 1D experiments, without the need of lengthy titrations with isotope labelled proteins – which represented a major bottleneck in drug discovery by NMR up to now. In CSAR exchange broadening is experimentally suppressed and calculated CSA tensors are used to obtain clean data which are proportional to the affinity of a ligand. Thus, ligands can be ranked seamlessly in an hour per ligand, using previously calculated CSA-values. This enables efficient drug discovery, even on difficult-to-express biomolecules.

THURSDAY, APRIL 28, 2022 - continued

12:10-12:35 **Metabolomics in Nutrition Research: current status and future opportunities**

Presenting Author: Lorraine Brennan

Lorraine Brennan (UCD)

Applications of metabolomics in nutrition research has increased in recent years and can be grouped into one of the following: (1) Identification of dietary biomarkers for single foods or for dietary patterns (2) Applications to dietary intervention studies to help understand metabolic alterations (3) Study of diet-related diseases and (4) Personalised/Precision Nutrition. With respect to dietary biomarkers there has been a proliferation of publications in this field: these biomarkers have the potential to act as objective measures of dietary intake thus overcoming some of the key issues with traditional assessment methods. To date, metabolomic profiling has been successful in identifying several putative biomarkers of food intake. Similarly, use of combination of biomarkers can be employed to study dietary patterns.

ThOD: Instrumentation, Hyperpolarization, & Eclectica 3 (parallel session)

10:45-12:35, Grand F

Daniel Raftery presiding

10:45-11:10 **Next-generation Equipment for Clinical-scale Parahydrogen Induced Polarization**

Presenting Author: Eduard Y Chekmenev

Eduard Chekmenev (Wayne State University)

Next-generation clinical-scale parahydrogen-based hyperpolarization instrumentation is presented to enable hyperpolarization of [1-13C]pyruvate and other 13C- and 15N- biocompatible contrast agents. Order-unity 13C polarization of [1-13C]pyruvate (and structurally similar biomolecules) in seconds becomes possible via SABRE in SHield Enables Alignment Transfer to Heteronuclei using this equipment relying on simultaneous exchange of parahydrogen and to-be-hyperpolarized substrate on metal complex. Moreover, 15N hyperpolarization of [15N3]metronidazole in excess of 20% is demonstrated using this hyperpolarizer. The device has a footprint of 18.5”x24”x18.5” with the cost of the off-shelf components of less than $15,000. We also present on the development and validation of handheld disposable container for temporary parahydrogen storage, transportation and utilization to enable parahydrogen-based hyperpolarization on sites that do not have parahydrogen-production infrastructure.

11:10-11:30 **Intrinsic atomic gradiometer for sensitive RF detection**

Presenting Author: Robert J Cooper

Robert Cooper (George Mason University); David W. Prescott (George Mason University); Karen L. Sauer (George Mason University)

Two minimally-shielded atomic magnetometers in a gradiometer configuration demonstrate an optical subtraction technique used to reduce common-mode interference up to a factor of 385. In addition mode, these magnetometers are able to achieve sub-femtoTesla sensitivity at frequencies close to 1 MHz. They are fundamentally limited by photon shot noise. Amidst the presence of a 5.2 pT common-mode interference, detection of two-orders-of-magnitude smaller signal is clearly shown. This technique bypasses instrumental limitations due to large interference signals saturating receive channels. This scheme is particularly useful for localized sources as they would produce gradient signals over the sensors, as from an NQR sample such as sodium nitrite.

11:30-11:50 **Automated High-Throughput Nuclear Magnetic Resonance Spectroscopy**

Presenting Author: Omar Nassar

Omar Nassar (Institute of Microstructure Technology IMT, Karlsruhe Institute of Technology KIT); Mazin Jouda (Institute of Microstructure Technology IMT, Karlsruhe Institute of Technology KIT); Dario Mager (Institute of Microstructure Technology IMT, Karlsruhe Institute of Technology KIT); Jan G. Korvink (Institute of Microstructure Technology IMT, Karlsruhe Institute of Technology KIT); Neil MacKinnon (Institute of Microstructure Technology IMT, Karlsruhe Institute of Technology KIT)

Enhancing NMR spectroscopy throughput is essential to open the door for this information-dense analytical technique to be used in prominent fields such as drug discovery, clinical analysis, chemical process monitoring, metabolomics, and food analysis. Flow-based NMR is a promising approach for enhancing NMR throughput. However, the relatively large dead volumes in the flow tubes feeding the NMR detectors are a standing challenge. In this contribution, we introduce a novel NMR probe head which possesses a microfluidic system that tracks the samples' position and velocity in a dual-phase flow and synchronizes the NMR acquisition to separate the samples' spectra. Fully automated NMR spectroscopy of nine different 120 µL samples could be achieved within 3.6 min.

THURSDAY, APRIL 28, 2022 - continued

11:50-12:10 **Decoupling of Spin Decoherence Paths near Zero Magnetic Field**

Presenting Author: Sven Bodenstedt

Sven Bodenstedt (ICFO - The Institute of Photonic Sciences); Denis Moll (Max Planck Institute for Multidisciplinary Sciences); Stefan Glöggler (Max Planck Institute for Multidisciplinary Sciences); Morgan W. Mitchell (ICFO - The Institute of Photonic Sciences); Michael C.D. Tayler (ICFO - The Institute of Photonic Sciences)

Proton-deuterium isotopic substitution is a tactic frequently used in high-field solution-state NMR. At low magnetic fields the advantages of 1H/2H substitution may, however, be outweighed by introduction of additional decoherence mechanisms. In this presentation, we theoretically and experimentally investigate the relaxation rates of 1H/2H and other dual-species spin system. By changing the effective symmetry of the spin Hamiltonian by using XY4 decoupling, we demonstrate that different contributions of individual interactions can be switched off. As an example, deuterium spin decoupling near zero field is used to increase extend the 1H T1 relaxation time by a factor of 2. We expect the approach to broaden the spectrum of hyperpolarized biomedical contrast-agent compounds and hyperpolarization procedures that are used near zero field.

12:10-12:35 **Hyperpolarization Enables Fast Reaction Monitoring of Diluted Systems**

Presenting Author: Kerstin Muennemann

Kerstin Muennemann (Technical University Kaiserslautern)

Reaction monitoring is mostly carried out on-line: the mixture that is to be analyzed is pumped through the NMR instrument, which is operated in flow mode. However, the analysis of fast-flowing liquids with NMR spectroscopy is challenging because short residence times in the magnetic field of the spectrometer result in inefficient polarization build-up and thus poor signal intensity. This is particularly problematic for compact NMR spectrometers, because of the small volumes available for prepolarization. Hyperpolarization methods like Overhauser Dynamic Nuclear Polarization (ODNP) and Parahydrogen Induced Polarization (PHIP) are well suited to overcome this problem because hyperpolarization build-up happens very rapidly and can be performed under continuous flow. This enables monitoring of fast reactions even in diluted systems.

LUNCH-ON-YOUR-OWN
12:35-14:00

POSTER SESSION
14:00-15:45
See Poster Listings at end of document

ThOE: Awards & Small Molecules 3: In-situ & Town Hall-Helium (plenary session)
16:00-17:50, Grand DE
Joanna R. Long and Sophia Hayes co-presiding

16:00-16:20 **Presentation of the following awards:** Anil Kumar Travel Award, Melanie Rosay Young Academic Support Program Awards, Manrao-Rastogi Travel Awards for Young Scientists; and the Ritchey Travel Award

16:20-16:45 **What Small Molecules Can Unlock About Food-Derived Endogenous Effects**

Presenting Author: Hanne Christine Bertram

Hanne Christine Bertram (Aarhus University)

“You become what you eat” is a popular phrase, reflecting that we have a belief in that what we eat affects our health. This connection between diet and human health is backed up by substantial evidence from observational studies showing an association between our dietary patterns, life style diseases and life span. An example is data from epidemiological surveys pointing at a high intake of red and processed meat increases the risk of certain types of cancer and mortality. Data will be presented with the aim of demonstrating how proton NMR spectroscopic analyses of small molecules in blood, urine or feces might give us hints about the molecular mechanisms behind potential associations between dietary items, dietary patterns and human health.

THURSDAY, APRIL 28, 2022 - continued

16:45-17:10 **New Developments in the ADEQUATE Family of NMR Pulse Sequences**

Presenting Author: R. Thomas Williamson

Robert (Thomas) Williamson (UNC Wilmington); Gary E. Martin (Seton Hall University); Ronald C. Crouch (JEOL Inc.); Jeffrey G. Raab (UNC Wilmington)

ADEQUATE experiments were first described in the mid-1990s. In the intervening two decades, there have been significant advances in NMR technology that have reduced sample requirements from the ~ 10 mg range to less than a milligram. In parallel with advances in NMR probe technology, progress in areas like non-uniform sampling, and new variants of the ADEQUATE experiments have enhanced the intrinsic value of these experiments and slowed the flow of incorrectly reported structures. In other novel applications, 19F-Detected versions of the ADEQUATE class of NMR pulse sequences have been developed that allow for the visualization and measurement of the 1JCC and nJCC coupling constants of fluorinated alkyl substances implicated in myriad of alarming environmental contaminations across the USA.

17:10-17:50 **ENC Town Hall & Helium Emergency Discussion**

BOAT BUILDING AT THE POOL!
18:15-19:30

Up to 50 people can sign up to be on boat building teams. Engineering and problem solviing skills needed! As are brave souls willing to captain the 'boat' built by your team! EVERYONE invited to spectate! A cash bar will be available.

Look for sign-up board near the conference registration.

TACOS & TUNES
19:30-22:00

Ticket required ($50). Purchase ticket online until April 26 at 12pm noon.

Tasty tacos and tunes from steel drum band will relax you this evening. One drink ticket included (soda, wine or beer). Cash bar also available.

FRIDAY, APRIL 29, 2022

EARLY AM: SPIN DYNAMICS (plenary session)
07:00-08:00

07:00-08:00 **Simulation Tutorial** Part 5 of 5: **SPATIAL ENCODING**

Presenter: Ilya Kuprov

FOA: Biomolecular Solids 3: Membrane Proteins, Enzymes, and Protein Folding (plenary session)
08:30-10:20, Grand DE
Leonard Mueller presiding

08:30-08:55 **Proton detected MAS-NMR of membrane proteins**

Presenting Author: Loren Andreas

Evgeny Nimerovsky (MPI for multidisciplinary sciences); Eszter Najbauer (MPI for multidisciplinary sciences); Kumar Movellan (MPI for multidisciplinary sciences); Kai Xue (MPI for multidisciplinary sciences); Marcel Forster (MPI for multidisciplinary sciences); Riza Dervisoglu (MPI for multidisciplinary sciences); Xizhou Zhang (MPI for multidisciplinary sciences); Karin Giller (MPI for multidisciplinary sciences); Andrei Leonov (MPI for multidisciplinary sciences); Kerstin Overkamp (MPI for multidisciplinary sciences); Melanie Wegstroth (MPI for multidisciplinary sciences); Christian Griesinger (MPI for multidisciplinary sciences); Stefan Becker (MPI for multidisciplinary sciences); Loren Andreas (MPI for multidisciplinary sciences)

Protons are ideal NMR reporters since they are abundant in biological materials and have high intrinsic sensitivity. The limiting factor for magic-angle spinning (MAS) NMR of solids has been the proton linewidth. The past 2 to 3 decades have witnessed a phenomenal improvement in proton spectral quality by (1) sample deuteration, (2) faster MAS, and (3) higher magnetic fields. Here we report recent developments in methodology and application to five membrane proteins: AlkL, M2, Opa60, hVDAC, and CitA. Our emphasis is on the use of particularly high magnetic field, which allows for highly resolved proton spectra to be recorded with 55 to 100 kHz MAS without the need for sample deuteration.

08:55-09:15 **Higher-Dimensionality Fast-MAS Solid-State NMR: An (Unexpectedly Viable) Weapon Against Peak Overlap**

Presenting Author: Rasmus Linser

Alexander Klein (TU Dortmund University); Suresh K. Vasa (TU Dortmund University); Rasmus Linser (TU Dortmund University)

Higher-dimensionality correlations have long existed in solution NMR of intrinsically disordered proteins to enable extraordinary dispersion, but they have been of limited benefit for globular proteins. Given the recent revolution of solid-state NMR by fast magic-angle spinning and proton detection, complex pulse sequences with high sensitivity have become possible. Here I will show how these developments, combined with latest non-uniform sampling and data reconstruction, enable assignment of a 2x 72 kDa enzyme and thus redefine the prospects of solid-state NMR for complex protein targets. I will specifically focus on five-dimensional fast-MAS approaches for backbone assignment as well as a five-dimensional side chain proton assignment experiment, recorded in a combined way with a four-dimensional intra-side chain correlation.

09:15-09:35 **Structural Basis for Regulation of a Human GPCR by Membrane Lipids Investigated by NMR Spectroscopy**

Presenting Author: Matthew Eddy

Naveen Thakur (University of Florida); Arka Ray (University of Florida); Niloofar Pour (University of Florida); Beining Jin (University of Florida); Liam Sharp (University of Delaware); Edward Lyman (University of Delaware); Anuradha Wijesekara (University of Florida); Alexander Duong (University of Florida); Matthew Eddy (University of Florida)

G protein-coupled receptors (GPCRs) are sensory membrane proteins comprising the largest class of “druggable” targets. We present NMR data that show membrane phospholipids and sterols control the function-related conformational equilibria of GPCRs with a magnitude comparable to or greater than the influence of drug efficacies. Using NMR spectroscopy in aqueous solutions, we investigated the structural basis for these observations in nanodiscs containing the human A2A adenosine receptor (A2AAR) and mixtures of zwitterionic and anionic lipids and cholesterol. 19F-NMR data of A2AAR in lipid nanodiscs containing mixtures of phospholipids and sterols showed striking synergies with ligand efficacies. Mechanisms for the lipid-dependent behavior were further investigated with judiciously selected mutagenesis, molecular dynamics simulations, and correlative cell-based signaling assays.

FRIDAY, APRIL 29, 2022 - continued

09:35-09:55 **Time-resolved DNP-enhanced solid-state NMR captures protein folding and oligomerization triggered by sub-millisecond negative temperature jumps**

Presenting Author: Blake Wilson

Blake Wilson (National Institute of Diabetes and Digestive and Kidney Diseases); Wai-Ming Yau (National Institute of Diabetes and Digestive and Kidney Diseases); Robert Tycko (National Institutes of Health)

Structural conversion processes are at the heart of many biophysical systems of interest. We demonstrate an approach using rapid temperature jumps from high temperatures, where biomolecular systems can be made disordered, to low temperatures, where ordered structures form. After a short evolution period, conversion is halted by rapid freezing. Low-temperature MAS-DNP-enhanced solid-state NMR is used to interrogate the conversion process, allowing quantitative, site-specific measurements of conversion kinetics. Rapidly cooling a solution of melittin—a 26 residue peptide found in bee venom—from 90 °C to 30 °C in <0.8 ms initiates a conversion from disordered monomers to ordered helical tetramers, which is tracked with solid-state NMR. Measurements show secondary structure formation occurs on the same timescale as oligomerization.

09:55-10:20 **Molecular mechanisms of multidrug efflux pumps**

Presenting Author: Nate Traaseth

Nate Traaseth (New York University)

Antibiotic resistance arises from multiple molecular mechanisms, including enzymatic breakdown of drugs, mutations of target proteins, reduced drug influx, and the activation of efflux pumps. The efflux mechanism by membrane protein transporters is one of the broadest resistance mechanisms, and is accomplished by active transport that reduces the internal drug concentration. In this seminar, our recent progress in studying efflux pumps from pathogenic organisms will be discussed, including structure, dynamics, and functional studies.

COFFEE BREAK
10:20-10:45

FOB: Theory and Computation 3: DNP, Automated Shimming, and Inverse Transforms (plenary session)
10:45-12:35, Grand DE
Robert Schurko presiding

10:45-11:10 **Probing minority Components in Solids by NMR Spectroscopy – from Polymer Defects in weathered Microplastics to Self-Aggregation of Polymer Additives**

Presenting Author: Juergen Senker

Kasper P. van der Zwan (Inorganic Chemistry, University of Bayreuth); Anika Mauel (Inorganic Chemistry, University of Bayreuth); Bjoern Poetzschner (Inorganic Chemistry, University of Bayreuth); Robert Staeglich (Inorganic Chemistry, University of Bayreuth); Tobias Kemnitzer (Inorganic Chemistry, University of Bayreuth); Wiebke Riedel (Physical and Theoretical Chemistry, Freie Universität Berlin); marcel Meinhart (Inorganic Chemistry, University of Bayreuth); Fabien Aussenac (Bruker BioSpin); Christian Reiter (Bruker Biospin); Thomas Risse (Physical and Theoretical Chemistry, Freie Universität Berlin); Caroline D. Keenan (Department of Chemistry and Biochemistry, Carson-Newman University); Gaël De Paëpe (CEA / Univ. Grenoble Alpes); Juergen Senker (Inorganic Chemistry, University of Bayreuth)

Chemical modifications and additions to polymers dramatically change the properties of the base polymers, even if these changes occur with minuscule proportions of only a few hundred ppm. Here we show, that techniques for sig-nal enhancement like SEOP and DNP can be used to overcome the sensitivity challenge of NMR spectroscopy and allow to characterize structural aspects and identify chemical functionalities for the minority components. We present strategies developed for probing self-aggregation of molecular polymer additives within polymer melts and for unrav-elling the abiotic degradation mechanisms of microplastic particles when weathered under environmental conditions. While polymer additives expand the application range of polymers, weathering of microplastic particles changes in-creases interactions with natural colloidal particles and living organisms in the environment.

FRIDAY, APRIL 29, 2022 - continued

11:10-11:30 **Predicting MAS-DNP properties of biradicals**

Presenting Author: Frederic Mentink-Vigier

Frederic Mentink-Vigier (National High Magnetic Field Laboratory, Florida State University)

The limited sensitivity of solid-state NMR makes studying systems with low concentration of the isotope of interest a challenge. To overcome this, bis-nitroxide radicals are commonly used to enhance nuclear polarization via Magic Angle Spinning Dynamic Nuclear Polarization (MAS-DNP) experiments. For the past ten years, we have developed a methodology that combines EPR, DFT, MD with a theoretical model to simulate large spin systems. This methodology can yield quantitative MAS-DNP simulations for standard systems as demonstrated with the bTurea (bcTol, AMUPol and bcTol-M) and AsymPol families. It reveals how slight modifications impact MAS-DNP performance. We also demonstrate that the performance of AsymPol-POK and cAsymPol-POK improves in proton dense media, bypassing the need to deuterate samples.

11:30-11:50 **Microwave-Free J-driven Dynamic Nuclear Polarization (MF-JDNP): A Proposal to Enhance the Sensitivity of Solution State NMR**

Presenting Author: Maria Grazia Concilio

Maria Grazia Concilio (Weizmann Institute of Sciences); Ilya Kuprov (University of Southampton); Lucio Frydman (Weizmann Institute of Science)

We have recently proposed J-driven Dynamic Nuclear Polarization (JDNP) for enhancing the sensitivity of solution-state NMR at high fields, and bypass some of the limitations faced by Overhauser DNP. JDNP relies on irradiating biradicals possessing an interelectron exchange coupling equal to the electron Larmor frequency, by high-frequency, high-power microwaves. This study proposes to bypass the heating and poor sample penetration challenges associated to such irradiation, with what we denominate as microwave-free J-DNP (MF-JDNP). MF-JDNP relies on shuttling the sample through the J-DNP condition; when coupled to the relatively long lifetimes envisioned for the singlet states arising in the strongly-coupled bi-electron system, this opens an opportunity to enhance NMR sensitivity at high magnetic fields –even in aqueous solutions- without irradiation.

11:50-12:10 **Deep regression with ensembles enables fast, first-order shimming in low-field NMR**

Presenting Author: Moritz Becker

Moritz Becker (Karlsruhe Institute of Technology); Mazin Jouda (Karlsruhe Institute of Technology); Anastasiya Kolchinskaya (Karlsruhe Institute of Technology); Jan G. Korvink (Karlsruhe Institute of Technology)

Shimming precedes most NMR acquisition procedures, and although it is predominantly a semi-automated procedure, it often requires repetition and experience, which might be cumbersome and time-consuming. We present a proof of concept that deep learning (DL) can simplify and accelerate the shimming problem. It is shown that DL can relate measured spectral shape to shim current specifications and thus rapidly predict three shim currents simultaneously, given only four input spectra. We also introduced a dataset that served as our DL training set, and allows inference of changes to 1H NMR signals depending on shim offsets. In-situ experiments of our method demonstrate a high success rate in spectral quality improvement for random shim distortions over different neural architectures and chemical substances.

12:10-12:35 **An Inverse Problems Framework for Magnetic Resonance Relaxometry and Macromolecular Mapping**

Presenting Author: Richard Spencer

Richard Spencer (National Institutes of Health)

Conventional MRI is a Fourier technique, with image reconstruction having the attractive property of being mathematically well-conditioned; noise in the data is transmitted to the image, but not magnified. In contrast, reconstruction in MR relaxometry is via the inverse Laplace transform, a form of the classically ill-posed problem of solving the Fredholm equation of the first kind. As a result, specialized methods must be undertaken to produce useful results. The inverse problems perspective has proven to be enormously fruitful in this setting. We will discuss this framework, and our applications to myelin mapping. The goals of our work are to improve the capacity of MR to evaluate tissue pathology and to develop methods for application to inverse problems more generally.

ADJOURN

63rd ENC PROGRAM (as of 14-April-2022)

Refer to Online Planner & Mobile App UPDATED PROGRAM. This file is for general reference only.

Odd-numbered posters present Mon and Wed; Even-numbered posters present Tues & Thurs.

Missing poster numbers represent late withdrawals.

POSTER TOPICS-OVERVIEW

POSTER 002

Water Accessibility Refinement of the Extended Structure of KirBac1.1

Presenting Author: Reza Amani

Complete Author List:
Reza Amani (Department of chemistry and biochemistry, Texas Tech University); Charles D. Schwieters (National Institutes of Health); Maryam Yekefallah (Department of chemistry and biochemistry, Texas Tech University); Isaac R. Eason (Department of chemistry and biochemistry, Texas Tech University); Benjamin J. Wylie (Department of chemistry and biochemistry, Texas Tech University)

NMR structures of membrane proteins are often hampered by poor chemical shift dispersion and internal dynamics which limit resolved distance restraints. Membrane protein water accessibility surface area is often investigated as a topological function via solid-state NMR. Here we leverage water-edited solid-state NMR measurements in simulated annealing calculations to better resolve membrane protein structure. We use solid-state NMR to measure water proximity and use this information to solve and refine the structure of KirBac1.1 using an updated version of Xplor-NIH. This is one of the largest SSNMR membrane protein structures yet reported (KirBac1.1, 148 kDa). All structural quality metrics indicate water accessibility restraints are a powerful way toward high quality structures of membrane proteins using NMR.

POSTER 003

Insight into the curvature control mechanism of the Rous sarcoma virus capsid protein assembly

Presenting Author: Bo Chen

Complete Author List:
Bo Chen (University of Central Florida); Tyrone Thames (University of Central Florida); Xin Qiao (University of Central Florida); Alexander Bryer (University of Delaware); Jaekyun Jeon (NIDDK, NIH); Peter Gorkov (National High Magnetic Field Laboratory); Ivan Hung (National High Magnetic Field Laboratory); Zhehong Gan (National High Magnetic Field Laboratory); Juan R. Perilla (University of Delaware)

The polymorphism of mature retroviral capsids is caused by insertion of twelve pentameric capsid proteins (CAs) into the hexameric lattice. Here we report the studies of the assembly of the 237-residue Rous sarcoma virus (RSV) CA, in its tubular assemblies and T=1 capsid, comprising entirely hexamers and pentamers, respectively. Atomistic models were obtained by combining ssNMR restraints with cryoEM density map. Similar residue specific intermolecular contacts are retained in both assemblies. SsNMR results identified twelve residues in distinct backbone structures in the tubular vs. pentameric assemblies, while the two domains of RSV CA undergo a 34 degree rotation around the flexible interdomain linker, to transform between the pentameric and hexameric assembly, without appreciable changes of the folding of individual domains.

POSTER 004

1H Detected MAS NMR of Human Voltage Dependent Anion Channel 1

Presenting Author: Edward P. Saliba

Complete Author List:
Edward Saliba (MIT); Ravi Shankar Palani (MIT); Choon Boon Cheong (Agency for Science, Technology, and Research); Robert G. Griffin (MIT)

Human voltage dependent anion channel 1 (hVDAC1) is the most abundant protein in the mitochondrial outer membrane. One of the most important ways that it fulfills this role is through its behavior as a highly efficient gate for the energy storage molecules ATP and NADH. The mechanism behind this gating, however, is not well understood. Presented are 1H detected multidimensional NMR spectra of hVDAC1 at 90 kHz of MAS in an 800 MHz magnet. Despite the relatively long amino acid sequence of hVDAC1 (283 residues), the 3 dimensional hCONH, hCANH, and hCCH spectra show site specific resolution. The assignment of these resonances will be an important step towards determining the gating mechanism of hVDAC1.

POSTER 005
A new antibiotic forms fibrils to destroy the bacterial membrane
Presenting Author: Rhythm Shukla

Complete Author List:
Rhythm Shukla (Utrecht University); Francesca Lavore (Utrecht University); Sourav Maity (RUG); Maik Derks (Utrecht University); Raj Kumar (Utrecht University); Alexandre MJJ Bonvin (Utrecht University); Dallas Hughes (Novobiotic Pharmaceuticals); Eefjan Breukink (Utrecht University); Kim Lewis (Northeastern University); Markus Weingarth (Utrecht University)

The discovery of the powerful antibiotic teixobactin (Nature, 2015) marked a new era for our combat against multi drug-resistant superbugs. While it is known that teixobactin blocks the cell wall synthesis by targeting Lipid II, structural data are scarce and were exclusively acquired in non-physiological media such as micelles.
Using a solid-state NMR approach, microscopy, and computational studies, here we succeeded to resolve the mode of action of teixobactin directly in native bacterial membranes. With advanced microscopy we show that sequestering of the target molecules in fibrils on membranes serves as an alternate killing mechanism. Our results explain the excellent bactericidal activity of teixobactin and provide crucial information on its mode of action for development of better antibiotics.

POSTER 007
Studying molecular changes at the cell / extracellular interface with Goldman-Shen experiments
Presenting Author: Thomas Kress

Complete Author List:
Thomas Kress (University of Cambridge); Melinda J. Duer (University of Cambridge)

Cell membrane interfaces are of paramount importance to sustain cell activity and life. Conventional ssNMR spectroscopy is a valuable tool to perform structural studies on biological tissues, yet it is not well-adapted to study interfaces, as is it a bulk-sensitive technique. In this work, we propose that spatial selectivity can be achieved with solid-state NMR using Goldman-Shen-like experiments. They rely on the T2(1H) selection of the cell membrane 1H magnetisation, and its transport to the region of interest via spin diffusion. We characterise the spin diffusion dynamics on 13C-enriched vascular smooth muscle cells (VSMCs), and present relevant control experiments to remove unwanted relaxation effects.

POSTER 008
Solid-state NMR Characterization of Lyophilized Formulations of Monoclonal Antibodies
Presenting Author: Luke W Arbogast

Complete Author List:
Jacqueline Perodeau (Department of Chemistry and Chemical Biology, Rutgers University); Andrew J. Nieuwkoop (Department of Chemistry and Chemical Biology, Rutgers University); Luke Arbogast (Biomolecular Measurement Division, National Institute of Standards and Technology)

With the introduction of monoclonal antibody (mAb)-based therapies for COVID-19 there is a renewed need for improved methods to assess their critical quality attributes, such as higher-order structure (HOS). Solution-state NMR has proven to be a promising method for such characterization, however, since many mAbs are prepared in lyophilized form, the ability to probe HOS in the solid-state is also important. We demonstrate the proof-of-concept for using solid-state NMR ^{1}H-^{13}C cross polarization (HC-CP) buildup for HOS characterization of lyophilized mAb formulations and show how Principal Component Analysis of HC-CP buildup spectra can

differentiate between lyophilized forms of the NISTmAb model system. Results support the use of solid-state NMR as a means to characterize solid-phase preparations of mAbs and other biotherapeutics.

POSTER 009
13C Natural Abundance Solid-State NMR and DNP of Plant Tissues and In-Vitro Synthesized Biomaterials
Presenting Author: Fabien Deligey

Complete Author List:
Fabien Deligey (Louisiana State University); Wancheng Zhao (Louisiana State University); Alex Kirui (Louisiana State University); Tracy B. Nixon (Pennsylvania State University); Daniel Cosgrove (Pennsylvania State University); Frederic Mentink-Vigier (National High Magnetic Field Laboratory, Florida State University); Tuo Wang (Louisiana State University)

Plant biomass is an important reservoir of renewable energy and understanding its complex structure is key to the improvement of biofuel production. We present three approaches using 13C natural abundance solid-state NMR and MAS-DNP for carbohydrate analysis. Assisted by spectral deconvolution, we establish a quantification method to detail the carbohydrate composition in onion cell walls. Enabled by DNP, we acquire high-resolution 2D 13C correlation spectra on unlabeled rice stems taken from the field, which allows us to resolve the variable conformations of cell wall polysaccharides. Combining MAS-DNP with CryoElectron Tomography, we visualize carbohydrate structure on both nano- and atomic-scales, with a demonstration on cellulose fibrils synthesized in-vitro through isolated proteins. Together, these strategies will facilitate high-resolution analysis of unlabeled biomaterials.

POSTER 010
Remodeling of the Fungal Cell Wall Structure by Antifungal Drug
Presenting Author: Malitha C Dickwella Widanage

Complete Author List:
Malitha Widanage (Louisiana State University); Frederic Mentink-Vigier (National High Magnetic Field Laboratory); Jean-Paul Latgé (Institut Pasteur); Ping Wang (Louisiana State University Health Sciences Center); Tuo Wang (Louisiana State University)

Recently, echinocandin drugs with reduced toxicity were developed to disrupt glucan synthesis and disrupt the integrity of the fungal cell walls by targeting their carbohydrate content. We employed solid-state NMR and living Aspergillus fumigatus cells to characterize the structural remodeling of fungal cell walls as induced by these antifungal drugs. The resulting cell walls exhibit uniquely high hydrophobicity and stiffness, as revealed by water-edited experiment and relaxation approaches, to resist external stress. The fungi also increased the content of chitin and produced new forms of α-glucans to compensate for the loss of β-glucan due to drug inhibition. These results will provide the structural foundation for designing future antifungals with improved efficacy.

POSTER 011
Binding Sites of a Positron Emission Tomography Imaging Agent in Alzheimer's β-Amyloid Fibrils from 19F Solid-State NMR
Presenting Author: Pu Duan

Complete Author List:
Pu Duan (Massachusetts Institute of Technology)

Positron emission tomography (PET) agents bind to amyloid fibrils, enabling early diagnosis of neurodegenerative disorders such as Alzheimer's disease. To understand the structural basis for PET binding to amyloid peptides, we investigate the binding sites of flutemetamol, a fluorine-containing PET tracer, in Aβ40 fibrils. Analytical HPLC and 19F NMR spectra show that flutemetamol binds Aβ40 fibrils with a stoichiometry of one ligand per 4-5 peptides. 1D and 2D 13C-19F and 1H-19F REDOR data reveal that three segments of the peptide lie the closest to the ligand. Distance-constrained docking indicates that these segments form multiple binding sites, and the ligand orientations at these sites are similar across multiple Aβ polymorphs.

POSTER 012
H-MAS
Presenting Author: Ago Samoson

Complete Author List:
Mai-Liis Org (Tallinn University of Technology); Ago Samoson (Tallinn University of Technology); Kalju Vanatalu (Tallinn University of Technology); Ats Kaldma (Tallinn University of Technology)

We have developed a 0.5mm spinning unit which can provide MAS speeds over 200 kHz and improves resolution of 1H in solids. High speeds are also beneficial for suppression of the first order quadrupolar interactions. Small rotor volume faciltiates a use of high-Q resonators for microwaves, reducing power requirements to the range of solid state amplifiers.

POSTER 014
Active site chemistry in a 206 kDa PLP enzyme enabled by DNP enhanced Solid State NMR Spectroscopy.
Presenting Author: Rittik K. Ghosh

Complete Author List:
Rittik Ghosh (Department of Biochemistry, University of California - Riverside, CA 92521); M. Luisa C. Nogueira (Department of Biochemistry and Molecular Biology, McKnight Brain Institute, National High Magnetic Field Laboratory, University of Florida, FL 32610.); Frederic M. Vigier (National High Magnetic Field Laboratory, Florida State University); Joanna R. Long (Department of Biochemistry and Molecular Biology, McKnight Brain Institute, National High Magnetic Field Laboratory, University of Florida, FL 32610.); Leonard J. Mueller (Department of Chemistry, University of California - Riverside, CA 92521)

Active site protonation states are critical for understanding the mechanisms of enzymatic transformations. However, protonation states are challenging to extract in large enzyme systems, such as Tyrosine Phenol Lyase (TPL), a 206 kDa pyridoxal-5'-phosphate (PLP) dependent enzyme. Here, we make use of DNP-enhanced solid-state NMR that enables the investigation of active site catalysis using site-specific substrate labels. For the TPL quinonoid intermediate, the combination of 1D 15N CPMAS and DNP enhanced 2D NCACX correlation spectroscopy allows us to determine the protonation states and unravel the catalytic mechanism via unique cofactor and active site chemical shift resonances. Through this process, we are able to observe the canonical quinonoid intermediate at atomic resolution under conditions of active catalysis.

POSTER 015
Three-Dimensional 13C Correlation Solid-State NMR for Carbohydrate Analysis
Presenting Author: S. Chandra Shekar

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Two and three dimensional (2D and 3D) solid state NMR have made major inroads in overcoming barriers to structural elucidation in isotopically (13C) enriched plant and fungal cell walls comprising mainly of complex carbohydrates. Yet, these efforts are partially hampered by heavily congested regions even in the 3D spectra. Replacing the two indirect single quantum (1Q) dimensions by an INADEQUATE 2Q-1Q dimensions, followed by a CORD mixing, eliminates body diagonal of the 3D cube and relieves congestion, ushering in dramatic resolution enhancement in plant and fungal cell wall spectra.

POSTER 016
Time-resolved DEER EPR and solid-state NMR afford kinetic and structural elucidation of substrate binding to Ca2+-ligated calmodulin
Presenting Author: Jaekyun Jeon

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Calmodulin regulates a range of target proteins in response to calcium concentration for numerous cellular/physiological functions. Upon binding with 4Ca2+, Calmodulin undergoes structural rearrangement that enables the target protein binding. The Calmodulin-binding domain peptide of Myosin Light Chain Kinase proteins (M13), for this study, is intrinsically disordered when unbound; forms a helical structure when bound with Calmodulin. Dynamics and structural details have been well studied, however, a kinetic description of the complex formation mechanism with proper time- and spatial resolutions have been elusive. Here, we implemented a recent millisecond time-resolved solid-state NMR approach combined with time-resolved and Tm-edited DEER EPR. Data afforded by both time-resolved approaches have revealed mechanisms comprising a bifurcating kinetic pathway with detailed structural insights of intermediates.

POSTER 017
Structural and Dynamics Studies of PIP3 in Lipid Bilayers Containing Phosphatidylserine and Calcium Cations
Presenting Author: Ashley D Bernstein

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Phosphatidylinositol phosphates (PIPs) are one class of signaling lipids that plays a significant role in membrane processes but are not well understood. By studying one of the PIPs, PIP3, in lipid bilayers, we aim to gain increased understanding of the structural and dynamics changes when PIP3 is exposed to phosphatidylserine and calcium cations. Using 1H and 31P MAS Solid-state NMR, we have observed dramatic changes in the sample upon each variation in sample. We seek to understand these changes and relate them to changes in the PIP3 head group structure and dynamics, perhaps showing clustering. Our intention is to use the insight gained into PIP3 behavior in lipid bilayers to better understand the mechanism of PIP3-protein binding regulation.

POSTER 018
DNP solid-state NMR to characterize Tau fibril structure.
Presenting Author: Riviere L. Gwladys

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Pathological aggregation of the protein tau into amyloid fibrils is a hallmark of different neurodegenerative diseases named tauopathies. Recently, ex vivo tau amyloid structures of all major tauopathies have been resolved by cryoEM. Interestingly, these structures are significantly different between diseases suggesting a critical interplay between the amyloid structure and the nature of the disease.
In order to gain insight into the factors that drive the formation of distinct tau amyloid structures, we study in vitro aggregated tau fibrils using solid-state NMR. The combination of MAS - DNP experiments with selective labeling allowed for measuring specific distances. Moreover, 1H detected fast-MAS experiments at 1.2 GHz provided unique insight into the structure of the tau fibrils.

POSTER 019
Visualize Fungal Cell Wall Organization Using Solid-State NMR, Functional Genomics, and Statistical Analysis
Presenting Author: Liyanage Devthilini Fernando

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The cell wall structure of four Aspergillus fumigatus mutants depleted of major structural polysaccharides in the intact cell walls were compared using high-resolution ssNMR spectroscopy. It revealed a rigid core formed by chitin, β-1,3-glucan, and α-1,3-glucan, with galactosaminogalactan galactomannan, and α-1,3-glucan present in the mobile phase. Also identified hydrophobic amino acids in alkali-insoluble fractions. In addition, we investigated the structural heterogeneity of chitin and chitosan in six fungal species using NMR chemical shift data, assisted by principal component analysis. The structure of chitin is found to be intrinsically heterogeneous, with unique fingerprints documented. Fungal chitin exhibits partial similarity to the α- and γ-chitin. The structural resistance of chitin to antifungal drugs and variation in salts concentrations was also revealed.

POSTER 020
NMR and Molecular Dynamics Investigations of Kindlin-2 Binding to Phosphatidylinositol Phosphates
Presenting Author: Andrew J Nieuwkoop

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Kindlin-2(K2) is a peripheral membrane protein regulated in part through its binding to phosphatidylinositol phosphates (PIPs). The Nieuwkoop lab is working to use NMR and molecular dynamics simulations to understand the properties of PIP containing lipid bilayers,

map the PIP binding sites of K2 PIP binding domains, and characterize the effects of PIP binding on the structure and dynamics of K2. 1H detected solid-state NMR experiments at 100+ kHz MAS are combined with 1H, 13C, and 31P detected experiments acquired at static, 15, and 40 kHz. All atom molecular dynamics simulations of the K2 PIP binding domains bound to a variety of PIP species and protonation states are used to drive questions and probe the importance of PIP protonation state.

POSTER 021
Distinct Pore-forming Conformation of Amyloid Beta Peptide Aβ1–42 in Membrane Environments
Presenting Author: Md Imran Khan

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The extracellular accumulation of fibrillar assemblies of amyloid beta (Ab) peptides in patients' brains is a hallmark of Alzheimer's disease. The interactions of lipid membrane and Ab peptides are known to further modulate the assembly and cytotoxicity of Ab peptides. However, there is no consensus regarding the effect of their interactions. In this work, we reconstitute Ab1-42 peptides in lipid bilayers emulating various important components in the cell membrane and apply solid state NMR (ssNMR) to characterize the resulting assembly structure. Our ssNMR results find Ab1-42 peptides in a distinct conformation from those observed previously, due to the presence of lipids, and provide essential structural restraints for Molecular dynamics simulations to establish a high-resolution model.

POSTER 022
The HRMAS Perspective on Bone Research: Evidence of Lipid Bilayers and Bone Resorption in Atlantic Herring Intermuscular Bones
Presenting Author: Hsin Wang

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T2 relaxation times of protons in bones cover 5 orders of magnitudes by MRI studies. There are mobile substructures among hard solid hydroxyapatite. The intermuscular bones (IB) of herring fish have no marrows, and adult IB contain more pore water. The INEPT of a small IB powder sample shows proton signals from long-chain fatty acids. T2 analysis along with water changes impacted by MAS suggests part of the lipids are phospholipid bilayers. By contrast, mobile water and oligopeptides with T2 ~ 20ms were detected in a large IB powder sample. They were confirmed by 1H-DIPSI-2-1H/13C-INEPT to be characteristic of collagen amino acids. This proves unequivocally the bone resorption process by NMR, which has not been reported before.

POSTER 023
Objective Assessment of Multidimensional Solid-State NMR Protein Spectra
Presenting Author: Benjamin D Harding

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Traditional assessment of multidimensional NMR spectraare subjective and time-consuming, especially for largemembrane proteins and fibrils in the solid state. Objective and timely analytics have potential tostrengthen rigor and reproducibility of procedures ranging from experimentalparameter optimizationup to all stages ofbiomolecular structure determination. Here we explore principal component analysis (PCA) techniques to assist in achieving these goals. We demonstrate PCA can be used to monitor spectrometer stabilityquantitatively, to provideconcise evaluationsof pulse sequence performance, toquantifythe regions of interest as a function of recoupling sequencetype and mixing time. Thus, theseutilitiesofferobjective approachesfor both data collection and processing, on the path towards real-time automation of instrument configuration and quality control.

POSTER 024
Determination of Histidine Protonation States in Proteins by Fast Magic Angle Spinning NMR
Presenting Author: Roman Zadorozhnyi

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Histidine residues play important roles in structure and functions of proteins depending on the ionization state of the imidazole ring. Solution and MAS NMR 13C, 15N and 1H chemical shifts are unique reporters of His protonation and tautomeric states. Here we present a fast MAS NMR approach for characterization of protonation and tautomeric states of His in proteins. Using 1H detected 2D heteronuclear correlation experiments with selective magnetization inversion techniques and TEDOR-based filters, we demonstrate for HIV-1 CACTD-SP1 in the microcrystalline state that these experiments permit unambiguous assignment of the protonation and tautomeric states of individual His residues. We anticipate that the approach presented herein will be useful for determination of His protonation states in large proteins and protein assemblies.

POSTER 025
DNP-Enhanced, Double-Quantum Filtered DARR/PDSD scheme to obtain 13C-13C Dipolar Correlation Spectra of Natural 13C Abundant Bone-Tissue Sample
Presenting Author: Sungsool Wi

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Our

POSTER exhibits a DNP-enhanced DQF DARR/PDSD scheme for obtaining 2D 13C-13C correlation spectra of the natural 13C abundant bone-tissue material. This scheme possesses a potential in obtaining pairwise 13C-13C distance information in the molecule because all the detected 13C-13C dipolar pairs are isolated probabilistically and, therefore, there are no dipolar truncation effect and relayed-fashion signal transfers involving multiple 13C sites. This scheme would be useful for the characterization of rigid biosolids or polymer materials in natural 13C abundant state.

POSTER 026
Accurate determination of N-H bond lengths in Watson-Crick and Hoogsteen A-T base pairs by DNP enhanced solid-state NMR
Presenting Author: Lakshmi Bhai N V

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Base pairings in double-stranded DNA usually exist in the canonical Watson-Crick(WC) conformation. However, various DNA complexes with protein and small molecules adopt an alternate Hoogsteen(HG) conformation. This conformational change occurs due to changes in hydrogen bonding between purines and pyrimidines. This study measures the hydrogen-bonded H-N bond length to detect changes in the structure of nucleotides due to conformation change. While detection of Hoogsteen base pairs is challenging for traditional structural biology techniques, previous studies from our lab have observed Watson-Crick and Hoogsteen base pairs with site-specific resolution with DNP Solid-State NMR. Therefore, DNP Solid-State NMR experiments, and SIMPSON spectral fitting was used to measure H-N dipolar coupling. The hydrogen-bonded H-N bond lengths were calculated from these dipolar coupling values.

POSTER 027
Histone Tails Dynamics in Chromatin Studied by High Resolution NMR Spectroscopy
Presenting Author: Nicole Gonzalez Salguero

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Despite their importance in DNA-templated process, histone tails have remained invisible to many traditional structural biology techniques.This has led us to use a combination of solution NMR and High-Resolution Magic-Angle spinning (HR-MAS) NMR to study histones H3 and H4 tails dynamics, focusing specifically on their interactions with nucleosomal DNA (nDNA) and linker DNA. We have used 147bp- single nucleosomes and 16mer nucleosome arrays (15-60bp linker DNA length) as our in vitro models. Our data suggests differences in conformational dynamics between nucleosomes and nucleosome arrays. This is due to transient interactions between the

tails and nucleosomal DNA or linker DNA. We have also found that linker DNA length has a weak influence in the dynamics of the H3 tails.

POSTER 028
Grafted sunflower trypsin inhibitor to inhibit protein-protein interaction: Constraints to convert multiple conformations to a major conformer.
Presenting Author: Seetharama Jois

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Peptides from CD2 were grafted onto SFTI to generate a bicyclic structure that is stable against enzymatic degradation. However, SFTI exhibits multiple conformations in solutions due to the presence of proline in the peptide structure. We incorporated a dibenzofuran moiety (DBF) in the peptide backbone structure instead of the Pro-Pro sequence to lock the grafted peptide into a major conformation in solution. The TOCSY spectra of the resulting peptide exhibited one set of major resonances for the SFTI peptide, whereas peptide with Pro-Pro sequence in the structure exhibited at least three or more sets of resonances for each amino acid in the NMR spectra. The peptide with DBF moiety was able to inhibit CD2-CD58 interaction at a submicromolar concentration in the cellular assay.

POSTER 029
Determining the 3D Structure and Dynamics of Mouse Notch1 Epidermal Growth Factor-Like Repeat 26 using Nuclear Magnetic Resonance Spectroscopy
Presenting Author: Kendra Jenkins

Complete Author List:
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Notch is a transmembrane protein that transmits signals between cells in direct contact and alters gene expression. The signaling pathway is important because it is involved in cell fate determination. The Notch1 extracellular domain has 36 epidermal growth factor-like (EGF) repeats that are responsible for ligand binding. In particular, EGF26 is known to be glycosylated catalyzed by the enzyme Pofut1 adding O-fucose and then recognized by Fringe, a Notch regulator that elongates O-fucose by adding N-acetylglucosamine (GlcNAc). We demonstrated that major changes in the chemical shift of amino acids are observed upon the addition of calcium resulting in a binding affinity of 2 mM indicating that calcium is needed to determine an accurate structure for EGF26.

POSTER 030
NMR Hydrogen Exchange Study of miR390a Precursor
Presenting Author: Ho-seong, Jin

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miRNAs originate from primary transcripts containing self-complementary hairpin structures that are initially processed to form 21~22-nt miR/miR* duplexes. In plants, primary transcripts with miRNA foldbacks (pri-miRNAs) are processed by the RNase-III like enzyme DICER-LIKE1 to generate miR/miR* duplex. The levels of mature miR390 influence the leaf number prior to flowering in the life cycle of plants. To understand the molecular mechanism of biogenesis of primary miR390a (pri-miR390a) to mature miR390, a NMR hydrogen exchange study was performed using model RNAs mimicking the cleavage site of wild-type and bulge-stabilizing mutant pri-miR390a constructs. Our results suggest that the stabilities of the two base-pairs at the cleavage site are essential for formation of the active conformation and for efficient processing of pri-miR390a.

POSTER 031
Unique Structural and Dynamic Properties of the HNH Nuclease in Thermophilic and Mesophilic Cas9 Revealed by NMR
Presenting Author: Helen B Belato

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CRISPR-Cas9 is a widely utilized biochemical tool. There are structural and dynamic changes occuring throughout Cas9 that are critical for target DNA recognition, unwinding, and cleavage. An understanding of the molecular motions guiding Cas9 and how this influences its mechanism are unknown. In this study we report on the structural and dynamic properties of the HNH domain of a thermostable Cas9 from Geobacillus stearothermophilus (GeoCas9). Here we show that GeoHNH motions are regulated by fast (ps-ns) timescale dynamics. Furthermore, when the residue with the highest flexibility on the ps-ns timescale is mutated in GeoHNH, its protein solubility, thermal stability, and dynamic profile are drastically distorted. Our results highlight key residues that play a role in giving GeoCas9 its thermophilic characteristic.

POSTER 032
Methodological Advances for the Characterisation of Human GPCRs by NMR Spectroscopy
Presenting Author: Philip Roessler

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G protein-coupled receptors (GPCR) are a pharmacologically important class of transmembrane proteins. The functioning of GPCRs is governed by their highly dynamic nature, which can be studied by NMR spectroscopy. However, most standard NMR techniques cannot be directly applied to GPCRs due to their large size and the necessity to use higher expression hosts.
We present new methods for NMR studies of GPCRs. We have established protocols for methyl and uniform 15N labelling in mammalian cells. Highly sensitive data acquisition on the resulting fully protonated samples is feasible using the novel XL-ALSOFAST-HMQC with delayed decoupling. Pseudocontact shifts were exploited for resonance assignment.
Using these methods, we characterise the conformational equilibrium of human β1-adrenergic receptor constructs in presence of diverse effectors.

POSTER 033
Interdomain Dynamics via Paramagnetic NMR on the Example of Calmodulin/Munc13-1
Presenting Author: Niels Karschin

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Paramagnetic NMR constraints are an excellent tool to study protein interdomain motion, but the interpretation is not always straightforward. On the example of the particularly flexible complex Calmodulin/Munc13-1 we present a new approach to characterize this motion with PCSs and RDCs. Using molecular mechanics, we have sampled the conformational space of the complex, and we have used a genetic algorithm to find ensembles that are in agreement with the data. We have used cross-validation and the Bayesian information criterion to determine the ideal ensemble size. This way, we were able to make an accurate, unambiguous, reproducible model of the interdomain motion of Calmodulin/Munc13-1.

POSTER 034
PEGylation of the Galectin-3 Carbohydrate Recognition Domain Creates a Kinetic Trap Protecting the Protein from Thermal Unfolding
Presenting Author: Emma Mulry

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Protein conjugation with synthetic polymers can improve their robustness, enabling new applications. However, we lack a detailed view of protein-polymer interactions that enable predictable protein stability due to limited experimental structural detail. We investigated the molecular interactions of PEG conjugated to the carbohydrate recognition domain of galectin-3 (Gal3C). Circular dichroism thermal melting experiments showed PEGylation resulted in increased stability, redirecting the unfolding pathway, and forming a stable intermediate. NMR revealed the global structure of Gal3C was preserved upon PEGylation, and a ‘shroud-like’ configuration of PEGylated Gal3C was supported by diffusion and hydrogen-to-deuterium exchange measurements. Gal3C-PEG interactions and unfolding could be influenced by replacing specific residues, suggesting the potential to rationally modify protein-PEG interactions while maintaining benefits of PEGylation.

POSTER 035
Improving the NMR spectral quality through different approaches to determine the 3D structure of the Chlamydial chaperone complex Scc4:Scc1
Presenting Author: Hemanthie C. Wickramasinghe

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Chlamydia trachomatis is responsible for the most sexually transmitted bacterial infections in the world. Scc4 is a bi-functional protein that acts as a type III secretion system chaperone and an RNA polymerase-binding protein. Scc4 forms a complex with Scc1 to bind the essential virulence factor, CopN. NMR structure determination of Scc4:Scc1 (33.5 kDa) is challenging due to signal overlap. In vitro and in vivo chain selective labeling methods were applied. In vivo labeling used the method by Mund et. al, LEGO-NMR, where the proteins are sequentially expressed in different media. The resulting Scc4:Scc1 complexes were not as stable as the complex produced by co-expression. Interpretation of these results suggests a model for Scc4 and Scc1 complex formation in Chlamydia trachomatis.

POSTER 036
Regulation of Human A2A Adenosine Receptor Conformational Dynamics by Endogenous Phospholipids
Presenting Author: Naveen Thakur

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G protein-coupled receptors (GPCRs) are sensory proteins that regulate many physiological processes and are targeted by one-third of FDA-approved drugs3. Using 19F-NMR in aqueous solutions, we investigated the role of anionic lipids on regulating the conformational dynamics of the human A2A Adenosine Receptor (A2AAR) in lipid nanodiscs. We explored the structural basis of the synergy of activation between drug efficacy and lipid composition by replacing specific charged amino acids with neutral residues at the receptor intracellular surface using 19F-NMR, MD simulations and cell signaling assays.

POSTER 037
Detecting the "undetectable": Functional protein motions in the hidden timescale window revealed by NMR relaxation measurements
Presenting Author: Supriya Pratihar

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Conformational flexibility dictates protein-function. Using 1H Extreme(E)-CPMG, we have detected binding relevant pincer-mode motion in ubiquitin for the first time. High-power RD revealed a very fast helix formation in the intrinsically disordered TAD domain of protein p53. The residues found to be involved in helix formation are known to bind protein MDM2 in tumor suppression during cancer progression. The supra-TauC gap in dynamics timescale spectrum can be closed with much higher B1 filed in 0.7 mm solenoid-coil in a MAS probe, which accommodates only 0.3 ul sample without possibilities of lock and gradients. To perform high-power RD on challenging proteins at low concentration, we are developing a solution-NMR probe, which allows 300 kHz B1 field on 40 ul sample volume.

POSTER 038
Translating Membrane Structure into Protein Function
Presenting Author: Yansheng Ye

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Atg3 is a key enzyme that catalyzes the conjugation of LC3 family proteins to phosphatidylethanolamine (PE) lipids during autophagy. Using NMR chemical shift perturbation, paramagnetic relaxation enhancement, and protein-lipid cross-saturation experiments we have discovered that in addition to the membrane-curvature dependent interaction of the N-terminal domain of human Atg3 (hAtg3), its catalytic region coincidently interact with the membrane. Our data indicate that hAtg3 uses a multifaceted membrane-association mechanism to bring substrates to proximity, optimally orients them, and promotes the transfer of LC3 from an LC3-hAtg3 intermediate to PEs in targeted membranes. Our results support an emerging concept that membrane geometry provides an essential cue for spatial regulation of autophagosome biogenesis during autophagy.

POSTER 039
New Insights into the Structure-Function Relationships of G Proteins Containing Disease-Associated Mutations
Presenting Author: Kara Anazia

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G proteins regulate intracellular signaling processes through their interactions with guanine nucleotides, GDP and GTP. In normal conditions, G protein functions are tightly controlled through interactions with partner proteins, G protein-coupled receptors (GPCRs). However, mutations in the α subunit of Gs disrupt this process, circumventing the usual control mechanisms and causing changes in Gs activity that result in serious illnesses including multiple cancers. We investigated the interactions of Gαs variants with nucleotides using saturation transfer difference nuclear magnetic resonance (STD-NMR) spectroscopy in aqueous solutions. Our results provide a better understanding of the effects of disease-causing mutations on nucleotide–G proteins interactions, and ultimately suggest approaches to target G proteins in the treatment of diseases.

POSTER 041
Membrane Interaction of the VPS37A N-terminal PUEV domain and its potential implications during phagophore closure
Presenting Author: Yansheng Ye

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The factors responsible for the phagophore closure during autophagy have been elusive. VPS37A as an ESCRT-I component is indispensable for phagophore sealing. Targeting of VPS37A to the phagophore is mediated by its N-terminal putative ubiquitin E2 variant (PUEV) domain. The sequence of this PUEV domain shares little homology with any proteins of known structure. Intriguingly, we have found that an N-terminal VPS37A construct (VPS37aN) that contains the PUEV domain, interacts with model membranes. We have identified two conserved regions that interact with the membrane (Figure 1A and 1B) and, moreover, the PUEV domain induces the formation of membrane high-order structure (Figure 1C). We are investigating a hypothesis that the PUEV domain recognizes and tethers highly curved membranes.

POSTER 042
Environmental influence on the pulmonary surfactant peptide SP-B1-25
Presenting Author: Maria Luiza Caldas Nogueira

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Pulmonary-surfactants (PS) attenuate alveolar surface tension. PS is synthesized by type-II-pneumocytes, stored as lamellar-bodies (LB), and secreted into the alveolar air space. Surfactant protein B is essential for the formation of LB, breathing, and newborn viability. In premature infants and lung, injured-old children the low PS leads to respiratory distress syndrome (RDS). Here we produced the 15N

labeled SP-B1-25 peptide and evaluate its behavior under low and high pH to understand how pH changes during the PS life cycle affect the trafficking of lipids

POSTER 043
Fully automated Lab-on-a-Chip system for studying protein-ligand interactions
Presenting Author: Sylwia Ostrowska Barker

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We demonstrate the capability of the NMR Lab-on-a-Chip system for the characterisation of protein-ligand interactions. A Protein-observed set of 1H-13C HSQC spectra of the SH3 domain of human Fyn protein in titration with the binding peptide from p85alpha is presented as a proof-of-principle validation of this system. This experiment was carried out in a fully automated fashion and required only minimal (approx. 30 uL) amount of protein sample. Focus on protein methyl groups can extend the application of this system into large molecular assemblies and protein-protein interactions. Alternatively, easy to implement adjustments to microfluidic sample manipulation can open the doors to studying multi-ligand equilibria.

POSTER 044
Structure of the ATP-Free Mre11-Rad50 DNA Damage Repair Complex Bound to DNA Substrates
Presenting Author: Mahtab Beikzadeh

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DNA double strand breaks are among the most harmful forms of DNA damage. The Mre11-Rad50-Nbs1 complex in eukaryotes and Mre11-Rad50 (MR) complex in prokaryotes plays a central role in the detection and repair of DSBs. Although there are some structural models available for MR complex bound to DNA, there are limitations in these structures. Therefore, our goal is to examine the MR complex bound to different DNA substrates using an integrated structural biology approach. We measured structural restraints for the P. furiosus MR complex bound to DNA using a variety of solution-based techniques (e.g., NMR spectroscopy, SAXS, and LRET) and used these data in subsequent structure calculations. Our models revealed novel modes of DNA binding for the ATP-free MR complex.

POSTER 045
Role of Cholesterol as an Allosteric Modulator for Human A2A Adenosine Receptor Conformational Dynamics
Presenting Author: Arka Prabha Ray

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Cholesterol is a well-recognized allosteric modulator of G protein-coupled receptors (GPCRs), but its mechanism of action is not well understood. We used 19F-NMR in aqueous solutions to investigate the structural basis for how cholesterol modulates the conformational dynamics of the human A2A Adenosine Receptor (A2AAR). We show that activation of A2AAR by small molecules also required the presence of cholesterol or other specific lipid compositions, and this allosteric mechanism is due to specific A2AAR-cholesterol interactions rather than through bulk membrane properties. Overall, our data point to the synergistic roles of drugs, cholesterol, and specific phospholipids in modulating the conformational dynamics of A2AAR.

POSTER 046
Characterizing Protein Folding Intermediates by Pressure-Jump NMR Spectroscopy
Presenting Author: Elahe Masoumzadeh

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Hydrostatic pressure can reversibly shift the thermodynamic equilibrium between folded and unfolded states, enabling experimental control over folding and unfolding. Using spectrometer-controlled hardware that performs rapid and repeatable pressure switching

within a sample cell, NMR spectroscopy can be utilized to record the residue-specific NMR signals immediately after a drop in hydrostatic pressure. We employed a pressure-sensitive mutant of ubiquitin (L50A) that shows multiple meta-stable folding intermediates. By identifying all resonances associated with the intermediate states that briefly develop when rapidly changing the sample pressure from 2 kbar to 1 bar, resonance assignment for multiple folding intermediates and parallel folding pathways were obtained. This work enables the first detailed structural folding study of a protein with multiple on-pathway intermediates.

POSTER 047
NMR as a Tool to Measure Activation of Ubiquitin and Ubiquitin-like Proteins by E1 Enzymes
Presenting Author: Westley Pawloski

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In the process of ubiquitination the small protein ubiquitin is ligated to a specific lysine on a substrate protein through an isopeptide linkage. The attachment process starts when ubiquitin-activating E1 enzyme binds and forms a reactive thioester with the C-terminus of ubiquitin. We have developed NMR-based methods to study this reaction, using a thiol-containing small molecule that displaces the enzyme and generates a new ubiquitin-thioester. The backbone amide of ubiquitin's C-terminal glycine has unique proton and nitrogen-15 chemical shifts, and a decay in its intensity upon thioesterification was monitored by 1D and 2D 1H-15N SoFAST-HMQC experiments to determine the reaction kinetics. Using this methodology we characterized the reaction velocities and KM values of E1 enzymes activating ubiquitin and ubiquitin-like proteins.

POSTER 048
Interactions Between Human GPCR Drugs and Membrane Lipids Studied by EPR and NMR Spectroscopy
Presenting Author: Beining Jin

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Many small molecules that interact with G protein-coupled receptors (GPCRs) and other membrane proteins are hydrophobic. A consequence of this is that small molecules interact with membrane mimetics and membranes within the cellular environment, but little is known about how membrane-drug interactions impact drug-receptor responses. To study this, we covalently attached a spin probe to an antagonist small molecule for the human A2A adenosine receptor and recorded EPR and NMR data to characterize drug-membrane interactions. We observe not only drug-receptor interactions but also significant drug-membrane interactions. Implications for the use of such molecules as sources of polarization for Dynamic Nuclear Polarization (DNP) SSNMR experiments will be discussed.

POSTER 049
Effect of SPIN N-terminus Stability on Affinity and Inhibition of Human MPO
Presenting Author: Nitin Mishra

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Staphylococcus aureus can evade killing within neutrophil phagosomes by producing a protein known as SPIN (Staphylococcal Peroxidase INhibitor). Structure/function analysis indicated that the triple-helical C-terminal domain of SPIN mediates binding to myeloperoxidase (MPO) while the N-terminal β-hairpin is required for inhibition. The SPIN N-terminus remains disordered in the unbound state due to the absence of intrinsic stabilizing features of its own. To further explore the structure/function relationships of the SPIN N-terminus, we introduced a disulfide in the SPIN N-terminus to trap it into the MPO-bound conformation. SPR analysis revealed that this mutant, called SPIN-cys (KD≈9.0nM), has a higher affinity for MPO than its WT (KD≈24.8nM). NMR studies were performed to calculate the solution structure of SPIN-cys and understand its dynamics.

POSTER 050
Characterization of the C-Terminal Segment of the S. Mutans Adhesin P1 by Solution NMR Spectroscopy
Presenting Author: Emily-Qingqing Peng

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Qingqing Peng (University of Florida); gwladys Riviere (DZNE); Joanna R. Long (University of Florida)

Streptococcus mutans is the virulent bacteria generating dental cavities. Previous studies indicate that the C-terminal region (C123, 51kDa) of the cell surface-localized adhesin P1 plays an important role in the formation of functional amyloids. We are characterizing the C123 amyloid formation by ssNMR. We are assigning the C123 backbone using solution NMR to identify key functional sites we can characterize in our ssNMR studies of the structural transition to amyloid. To assist in our assignment of C123, we are also assigning the C3 domain (19.5 kDa).

POSTER 051
Compressibility, Internal Motion and Conformational Entropy in the Thermodynamics of Molecular Recognition by Proteins
Presenting Author: Josh Wand

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The thermodynamics of molecular recognition is a central determinant of complex biochemistry. Recently, a dynamical proxy based on NMR-relaxation has revealed a richness for conformational entropy in the thermodynamics of ligand binding. We report the pressure-dependence of fast internal motion within the ribonuclease barnase and its complex with barstar and that protein dynamics are conserved along the pressure-binding thermodynamic cycle. The femtomolar affinity of the barnase-barstar complex exists despite a penalty by -TΔSconf of +11.7 kJ/mol at ambient pressure. At high pressure, however, the overall change in side chain dynamics is zero and binding occurs with no conformational entropy penalty. The pressure sensitivity clusters in response to both pressure and binding and a striking structural segregation of dynamics is observed.

POSTER 052
Conformational Transitions in Yeast Chorismate Mutase Important for Allosteric Regulation as Identified by Nuclear Magnetic Resonance Spectroscopy
Presenting Author: David D Boehr

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The chorismate mutase from Saccharomyces cerevisiae (ScCM), a key enzyme in the biosynthesis of aromatic amino acids, is allosterically activated and inhibited by tryptophan and tyrosine, respectively. Here we used NMR relaxation dispersion experiments to understand the conformational fluctuations on the microsecond-to-millisecond timescale that occur in ScCM. In the absence of allosteric effectors, ScCM did not exclusively exchange between T and R conformations, suggesting that the two-state MWC model is insufficient to explain conformational dynamics. Addition of tyrosine led to the quenching of much of the motion on this timescale, while new motions were identified in the presence of tryptophan. These new motions are consistent with conformational fluctuations into an alternative conformation that may be important for enzyme activity.

POSTER 053
Perfect TOCSY: 2–3 Fold Signal Enhancement Paves the Way for Assignments of Large Biomolecules
Presenting Author: Arnelle Löbbert

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Assignment of side-chain resonances remains a bottleneck for the investigation of large biomolecules. Hence, it is highly important to maximize signal intensity in assignment experiments.
We present two sensitivity improved 3D-CC-TOCSY experiments which benefit from a perfect echo (PE) element recovering signal losses arising from homonuclear J(CC) coupling. By suppressing the splitting from J(CC) couplings, narrow and intense singlet lines are obtained.
Signal intensities in the PE-HC(C)H-TOCSY are up to 2-fold enhanced compared to its non-PE counterpart, demonstrated for GB1 (8 kDa) and NanoBody80 (14 kDa).
For the PE-(H)C(CCO)NH-TOCSY, a combination of the PE element with a direct Hartmann-Hahn transfer from side chains to carbonyls leads to an even larger sensitivity increase as shown for MBP (43 kDa).

POSTER 054

Structural insights into GPCR incorporated into nanodiscs
Presenting Author: Nina Kubatova

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G protein-coupled receptors (GPCRs) belong to the largest transmembrane protein family in eukaryotes that play a vital role in intracellular signaling pathways and control various physiological processes. With the exception of class C receptors, there is no conclusive evidence of the oligomerization state of GPCRs. Using nanodiscs with higher radius, we incorporated several turkey β1-adrenergic receptor molecules into a single nanodisc. The distances obtained from the double electron–electron resonance (DEER) EPR spectroscopy provided insights into the oligomerization states of the GPCR in a native-like environment.

POSTER 055
UBact Dynamics and Binding to ARC ATPase Measured by 15N Relaxation
Presenting Author: Steven M. Bonn

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Proteasomes are ubiquitous across all domains of life. Proteasomal degradation is signaled by attachment of small modifier proteins such as ubiquitin in eukaryotes, SAMP in archaea, and Pup in certain bacteria. We have expressed and purified a new putative modifier protein, UBact, obtained a nearly-
Complete backbone NMR resonance assignment and characterized its dynamics. UBact is disordered in solution, similar to Pup, and in contrast to ubiquitin and other ubiquitin-like proteins. We have also characterized its binding to the putative hexameric proteasomal receptor in the UBact operon using the change in 15N relaxation rates upon binding. This work represents the first exploration of the UBact operon, and illustrates the use of spin relaxation as a robust method for measuring binding.

POSTER 056
Dynamic Structural Biology of Hyperactive Recurrent Cancer Mutations of NSDs
Presenting Author: Vladlena Kharchenko

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The nuclear receptor-binding SET domain-containing NSD family comprises selective H3K36 dimethyltransferases. The upregulation and recurrent hyperactive missense mutations of NSD enzymes are implicated in oncogenesis and chromatin regulation. Our studies demonstrated differences in functional dynamics between wild-type NSDs and their oncogenic hyperactive variants. NSD3SET (T1232A) showed the widespread mobility changes in the catalytic domain and regulatory loop. In the case of the E1099K mutation of the NSD2SET, activation of slow segmental motions across the SET domain leads to higher turnover rates of the SAM cofactor, causing increased H3K36me2 mark leading to oncogenesis. Identified structural dynamics landscape sheds light on the missense mutation-driven hyperactivity origins paving the way to target NSDs selectively

POSTER 057
Tractable approaches for solution NMR of glycoproteins
Presenting Author: Adam W Barb

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A conservative estimate reveals 22% of the human proteome is modified by a single type of carbohydrate, asparagine(N)-linked glycans, with the average protein receiving three glycans. These proteins are predominantly found at the cell surface or secreted, and the N-glycan modifications affect ligand binding, folding, stability, oligomerization, susceptibility to proteases and many other properties. Traditional expression hosts including E. coli do not synthesize N-glycan modifications, and yeast N-glycans contain features conserved in mammals but processing differs substantially. Here we present two strategies for expressing isotope-labeled N-glycoproteins using human cells and S. cerevisiae. We also present solution NMR measurements of the immunoglobulin G Fc, a 50 kDa dimer with one N-glycan, and the Fc gamma receptor 3a with five N-glycans.

POSTER 058
Characterization of structural features driving promiscuous ligand binding in GMCSF
Presenting Author: Jennifer Cui

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Granulocyte Macrophage Colony Stimulating Factor (GMCSF) is a cytokine important for the growth and differentiation of granulocytes and macrophages in humans. GMCSF participates in both protective and pathogenic biological processes in various immune-dysfunctional syndromes. For its immune function, GMCSF is administered clinically as an immuno-stimulatory agent. Previously, we show that three histidine residues modulators of structural perturbation in GMCSF. Our objective was to characterise contributions of each histidine using NMR and the driving forces of these contributions. We investigate this through mutagenesis of critical histidine residues using amino acids of various physical and chemical properties.

POSTER 059
Molecular Structures of Lanthanide-Binding Peptides for Recovery of Rare Earth Elements
Presenting Author: Surabh KT

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Rare earth elements play a pivotal role as indispensable components of state-of-the-art electronics. However, lanthanide elements are challenging to separate and recycle, largely due to their similar sizes and ionic charge densities. Recently, peptide surfactants that incorporate lanthanide binding tags (LBTs) have emerged as a green recovery method for rare earth elements. These peptide surfactants contain hydrophobic sequences for recovery at the air-water interface by foam froth floatation. However, to design peptides that bind specific elements, it is essential to understand how the local structures of the binding loop correlate with lanthanide ion selectivity. Herein, we elucidate the structures of two lanthanide-binding peptides (LBT3 & LBT1LLA), and the conformational changes associated with lanthanum complexation, through multi-dimensional solution-state biomolecular NMR methods.

POSTER 060
A structurally preserved allosteric site in the MIF superfamily affects enzymatic activity and CD74 activation in D-dopachrome tautomerase
Presenting Author: Emily Chen

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Macrophage migration inhibitory factor (MIF) and its homolog MIF-2 are cytokine-like proteins with critical roles in immunomodulation that are upregulated in various inflammatory diseases. While their potential as drug targets led to the creation of commercial MIF inhibitors, molecular level detail about MIF protein structure and dynamics are essential for effective therapeutic design. MIF contains a dynamic relay connecting the catalytic site to an allosteric site. We identify a comparable network in MIF-2 showing with nuclear magnetic resonance spectroscopy that dynamic communication is preserved in MIF-2 despite differences in primary sequence. We further characterize the MIF-2 pathway from the allosteric site to CD74 activating residues, defining a
Complete network that can be methodically targeted for functional control of the protein.

POSTER 061
Target DNA recognition of human transcription factor, MEIS1 studied by NMR
Presenting Author: Seo-Ree Choi

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Myeloid ecotropic viral integration site-1 (MEIS1) is a human transcription factor and plays an important role in myeloid leukemia. MEIS1 contains a three amino acids loop extension (TALE) homeobox domain (HD) and specifically binds to a common DNA sequence, 5'-TGACA-3'.

In this study, we performed NMR experiments on the complexes of TALE-HD of human MEIS1 (MEIS1-HD) with a decamer DNA duplex containing its consensus sequence. We prepared the mutant DNA duplexes containing a single base-pair substitution as well as double or triple substitution mutants and compared the MEIS1-HD-DNA interactions using HSQC titrations. This study provides key structural features of the MEIS-HD-DNA complex and the information about the molecular mechanism of target DNA recognition of the MEIS1.

POSTER 062
NMR study on the interaction of human HoxA1 with DNA
Presenting Author: Hyebin Ahn

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HoxA1 is one of homeodomain proteins highly conserved 60 amino acids. Although the second and third residues are conserved as Lys or Arg in almost all HOX proteins, the HoxA1 has residues Asn and Ala. In order to understand molecular mechanism of DNA recognition of HoxA1, we have performed NMR experiments on the homeodomain of HoxA1 (HoxA1-HD) complexed with consensus DNA and its mutant DNA duplexes. In addition, we prepared HoxA1-KR mutant and compared its structural feature in a complex with those of HoxA1-HD complex. We also determined the thermodynamic parameters for each DNA binding of HoxA1-HD and HoxA1-KR mutant using ITC. Our study provides an insight into the role of residues N2 and A3 during DNA recognition of HoxA1.

POSTER 063
NMR study on the interaction of human Pbx4 with DNA
Presenting Author: Youyeon Go

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PBX4 activates the ephA8 transcription and controls eye movement. PBX4 contains a three amino acids loop extension homeobox domain and specifically binds to a common DNA sequence, 5'-TGAT-3'.
In this study, we performed heteronuclear single-quantum correlation titrations on complexes of PBX4-EX and PBX4-HD with DNA duplex at various DNA-to-protein molar ratios. We also performed the 15N relaxation dispersion to study the kinetics of target-specific DNA binding of PBX4. We also determined the thermodynamic parameters for DNA binding of PBX4-EX and PBX4-HD using Isothermal titration calorimetry. This study provides insight into the structural role of the α4 helix of PBX4 protein during its target DNA recognition.

POSTER 064
Comparison of Z-DNA and Z-RNA binding mode of the P193A mutant of human ADAR1-Zα protein
Presenting Author: JUHEELIM

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JUHEE LIM (GNU); Joon-Hwa Lee (GNU)

HUman ADAR1 deaminates adenine in pre-mRNA to yield inosine (I), which codes as a guanine residue in mRNA. The Zα domains of human ADAR1 (ZαADAR1) preferentially binds to Z-DNA, rather than B-DNA, with high binding affinity. The substitution of P193, which located in β-hairpin, to A caused Aicardi -Goutières syndrome. In this study, we performed heteronuclear single quantum correlation (HSQC) titrations on complexes of the P193A mutant of ZαADAR1 with DNA or RNA duplexes at various DNA-to-protein molar ratios. Comparison of these results provides the structural information to explain the origin of Aicardi-Goutières syndrome.

POSTER 065
Protocol for Conformer Generation of Macrocyclic Drug Leads using NMR-Restraints
Presenting Author: Luciano Mueller

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Conformationally constrained macrocyclic peptidomimetic compounds offer an attractive venue for the design of orally active inhibitors of protein:protein interactions. Generation of 3D-conformers of macrocyclic drug candidates in solution permits testing the hypothesis that conformers of optimized compounds may approximate the target protein bound conformer. We will present results of NMR derived 3D-conformers of a drug lead whose crystal structure is known in target protein bound state. Our protocol comprises of first optimizing solvent conditions, followed by manual resonance assignments in SPARKY using a standard suite of 2D-spectra. NOE-peak lists exported from SPARKY are automatically assigned and upper bound restraints are generated in CYANA. Further minimizations and final optimized 3D-conformers are generated using the “forcegen”-protocol (Biopharmics, LLC.)

POSTER 066
NMR study of salt dependence of Dlx3-DNA interaction
Presenting Author: Juyeon Son

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The homeodomain transcription factor distal-less homeobox 3 gene (DLX3-HD) is required for hair, tooth and skeletal development. DLX3-HD specifically binds to a common DNA sequence, 5’-TAATTG-3’.
In this study, we performed NMR experiments on the complexes of the DLX3-HD with a decamer DNA duplex containing its consensus sequence. We present the binding affinity of the DLX3-HD for DNA using HSQC titrations at various salt concentration conditions. Also, hydrogen exchange rate constants of the imino protons for the DNA and DLX3-HD-DNA complex were measured by water magnetization experiment. This result provides structural features of the DLX3-HD-DNA complex and the mechanism of target DNA recognition of DLX3 transcription factors.

POSTER 067
pH-Dependent Aggregation of the Pmel17 Repeat Domain Characterized by Paramagnetic Relaxation Enhancement
Presenting Author: Daniel Morris

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In studying amyloid-forming proteins, it is imperative to determine the monomer conformational dynamics that precede fibrillation. This study utilizes NMR and the paramagnetic relaxation enhancement (PRE) effect to observe monomer properties of the repeat domain (RPT) from a human functional amyloid, premelanosomal protein (Pmel17). RPT is generated through Pmel17 post-translational processing during melanosome maturation, where melanin biogenesis occurs. At acidic melanosomal pH, RPT self-assembles into amyloid fibrils, functioning as a scaffold for melanin deposition. Here, we report solution conformations of the short (sRPT) isoform, which has been demonstrated to be a fibrillation nucleator. Intramolecular PRE suggest sRPT has a local conformation which protects residues associated with the amyloid core and that sRPT slowly depopulates from this conformation at low pH.

POSTER 068
Structural Characterization of La Related Proteins
Presenting Author: Blaine H. Gordon

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La Related Proteins are a superfamily of RNA binding protein whose homology is dictated by a highly conserved La domain and a Ribonucleotide Recognition Motif . Following the discovery and structural characterization of LARP3, it was suggested that the La/RRM pair was the minimum required unit for specific RNA binding function. While this holds true for certain LARPs, LARP1 and LARP6 have recently been shown to bind their RNA substrates with high affinity using only the La domain. Further, we've discovered a functional, unstructured region in LARP1. Our goal is to structurally and biochemically characterize the LARP1 La domain and the newly reported IUR together and separately to understand the role each plays in the regulation of mature, TOP-containing mRNA.

POSTER 069
Porosity and Saturation Measurement in Shales using NMR at Varying Pressures
Presenting Author: Stacey Althaus

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NMR has become a favorable non-destructive technique to determine porosity and hydrocarbon and water saturation in shales. Samples were saturated with Isopar at varying pressures and measured via low field NMR to determine the porosity and effective saturation. 1H CPMG T2 measurements were performed to assess the fluid quantity. The samples were found to be fully saturated after 4000 PSI pressure saturation. The fluid quantity was then converted to a porosity and was found to agree with industry standard methods. To further investigate the fluids in the samples, 2D T1-T2 measurements were performed using an inversion recovery-CPMG method. The better resolution given by two dimensions allows for the components to be separated and quantified individually.

POSTER 070
Pulse-Program-Controlled Rheo NMR: Enabling the Measurement of Liquid Crystal Phase Transition Kinetics and the Molecular Alignment below the Critical Concentration
Presenting Author: Burkhard Luy

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An apparatus for pulse-program-controlled rheological NMR-with up to 50 Hz spinning speed is introduced. We show its application to the widely known chiral liquid crystalline alignment medium PBLG with surprising results: at the LC phase transition deuterium NMR of the solvent proves the presence of coexisting phases formed with different time constants. In addition, tilted alignment is observed upon application of shear forces, which persists even at concentrations significantly below the conventional phase transition. We demonstrate that this very weak rheological alignment is of utmost interest for measuring RDCs of small molecules

POSTER 071
HOmonucleaR Recoupling by hEteroNuclear DecOUplingS (HORRENDOUS) NMR: Establishing correlations in solution-state NMR by reinstating non-secular J-terms
Presenting Author: Mihajlo Novakovic

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TOCSY is a well-established solution-state NMR experiment often complicated by high-power demands. This study explores an approach termed HOmonucleaR Recoupling by hEteroNuclear DecOUplingS (HORRENDOUS) that can achieve homonuclear correlations based on their J-couplings without pulsing on the nuclei to be correlated. The idea follows from considering H-C1-C2 three-spin system with J-couplings only active among directly-bonded spins. Nutation field w1H applied on C1-bound proton will introduce a modulation in the JCH coupling, that partially reinstates non-secular C1–C2 JCC flip-flop terms. Namely, 1H irradiation creates C1 decoupling sidebands and when these overlap the C2 spectral line efficient C1->C2 transfer can occur. These principles are demonstrated experimentally with different samples. Full theoretical description and possible applications of this method will be presented.

POSTER 072
Creating a digital archive of Barry Shapiro's NMR Newsletters
Presenting Author: Clemens Anklin

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We have undertaken the task of making the treasure trove of information that is contained in 516 issues of the MellonNMR, IITNMR, TAMUNMR and just NMR Newsletter available to past, current and future generations of practitioners of the art of magnetic resonance. This collection of groundbreaking, informative and sometimes whimsical information will be hosted on the ISMAR webiste.
Around the time of presentation of this work we hope to have near 300 issues done.
We are not in possession of an
Complete collection and are asking the community to help us make it whole.

POSTER 073
Overhauser DNP Solvent Dynamics Measurements of Binary Mixtures

Presenting Author: Timothy Keller

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ODNP is a technique capable of measuring local water/solvent dynamics. Recent instrumentation developments have allowed us to perform high resolution ODNP experiments at a polarizing field of 0.35 T (15 MHz). We have been able to achieve a linewidth of < 2 Hz (≤ 0.2 ppm) and demonstrate JRES on a sample of ethyl crotonate in a compact benchtop solution-state DNP-NMR spectrometer. The improved resolution introduces the possibility of performing chemical shift resolved solvent dynamics measurements for more complicated systems. As a model system, we use a binary mixture of water and acetonitrile and characterize the solvent dynamics of different molar ratios. Solvent dynamics measurements with free TEMPOL in solution and immobilized TEMPO on silica gel will be discussed.

POSTER 074
Disposable and Handheld Clinical-scale HP Propane Hyperpolarizer
Presenting Author: Nuwandi M Ariyasingha

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We present temporary parahydrogen gas storage in commercial disposable cheap (<$1) containers to enable on-site hyperpolarization of PHIP and SABRE molecules. Furthermore, parahydrogen:propylene gas mixture is stored for days in this handheld container to produce a clinical dose (>1L) of proton-hyperpolarized gas by performing the mixture ejection through disposable heterogeneous reactor. The feasibility of ultra-fast 2D EPI, GRE, and FIESTA imaging of HP propane produced using our hyperpolarizer at remote site has been demonstrated. This pilot study bodes well for near-future in vivo validation of this prospective inhalable contrast agent for functional lung imaging applications. All-in-all, the new instrumentation presented here enabled low-cost on-demand production of HP propane using a handheld device, and imaging using conventional clinical MRI scanners

POSTER 075
Hyperpolarization Ad-Infinitum: A Closed-Loop, Continuous-Flow System for Hyperpolarization of Catalyst-Free Metabolites via Heterogeneous Catalysis
Presenting Author: Yunpu Zhao

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Side-arm-hydrogenation (SAH) has extended the reach of parahydrogen (pH2) enhanced NMR to the hyperpolarization of metabolites such as pyruvate and acetate that are not directly producible by hydrogenation. Here we report a closed-loop continuous flow system, which enables perpetual recycling of unreacted liquid substrate reactant. The system consists of an HPLC pump, a liquid reservoir and pH2 gas bubbler, a tube in tube gas permeable membrane assembly for removal of nH2, and a packed-bed catalytic reactor. A continuous stream of hyperpolarized allyl acetate was maintained for over 20 minutes of re-circulation, with signal enhancements of up to 358 using 99% pH2. These promising results demonstrate the feasibility of performing systematic studies to optimize polarization yield and polarization transfer to heteronuclei.

POSTER 076
Temperature-Ramped Batch-Mode Spin Exchange Optical Pumping of Xenon-129 using 3rd-generation Automated XeUS Hyperpolarizer
Presenting Author: Md Raduanul Haque Chowdhury

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The clinical-scale batch-mode generation-3 XeUS hyperpolarizer device can perform hyperpolarization of 129Xe via Spin-Exchange optical pumping (SEOP) method safely at a temperature range of 60-95 °C. High-temperature SEOP is associated with a faster rate of polarization buildup but yields a lower %PXe. We demonstrate a hybrid temperature-ramp approach of batch-mode SEOP, which takes advantage of both high-temperature fast buildup rates and maximum %PXe at lower polarization temperature. This ramp approach (95-85 °C) yields %PXe = 39.8%, whereas the 95 °C polarization process produces %PXe = 35.9% in the same period of time of 42 minutes. We have also tested the 129Xe polarization retention after hyperpolarized gas ejection in an external phantom using ex-situ polarimetry of 47.5 mT portable MRI.

POSTER 077
Synthesis and 15N Hyperpolarization of Fluoro-[15N3]metronidazole via SABRE-SHEATH
Presenting Author: Mohammad Shah Hafez Kabir

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We performed ab initio calculations of 15N and 19F chemical shifts for fluoro-[15N3]metronidazole and its metabolic products due reduction in hypoxic conditions. The 15N chemical shifts exhibit a wide range of changes due to stepwise reduction, and we propose employing 15N chemical shifts for in vivo hypoxia sensing. In our present study, we have synthesized and hyperpolarized fluoro-[15N3]metronidazole using spin-relayed Signal Amplification By Reversible Exchange in SHield Enabled Alignment Transfer hyperpolarization method. All the three 15N sites of fluoro-[15N3]metronidazole have shown 15N polarization from 4.2% to 6.2%. Since direct 15N detection has poor NMR sensitivity even in hyperpolarized state, we explore the idea of 19F readout of 15N-hyperpolarized metabolites to improve the detection sensitivity.

POSTER 078
Hyperpolarized NMR of labile protein side chains
Presenting Author: Ludovica Martina Epasto

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Recently, dynamic nuclear polarization (DNP) became very popular as a hyperpolarization technique, used to boost signal intensities in NMR. Despite its wide applications in the field of protein NMR, the mechanism of polarization occurring during DNP is still not clear. In this study, we elucidate the mechanisms contributing to the transfer of polarization from the water to the target. Using arginine and poly-aspartate as molecular models, and employing molecular dynamic simulations, we describe the contributions of direct nuclear Overhauser effect (NOE) between water and target, exchange-relayed NOEs, and proton exchange on biomacromolecule. According to our results, amino acids with labile side-chain protons show particularly strong signal enhancements when dissolved in hyperpolarized water.

POSTER 079
Pulsed SABRE-SHEATH Improves Heteronuclear Polarization of [1-13C]pyruvate and [15N3]metronidazole
Presenting Author: Shiraz Nantogma

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Signal Amplification By Reversible Exchange in SHield Enables Alignment Transfer to Heteronuclei (SABRE-SHEATH) to provide low-cost alternative for hyperpolarization of biomolecules. We investigate the potency of recently developed pulsed SABRE-SHEATH approaches to improve heteronuclear polarization of hyperpolarized [1-13C]pyruvate and [15N3]metronidazole. We systematically optimized four experimental parameters of the square pulse applied during parahydrogen bubbling in microtesla magnetic field in our

new SABRE-SHEATH hyperpolarizer device. Our results show that pulsed SABRE-SHEATH can substantially improve 15N polarization of [15N3]metronidazole from 13.7% to 20.7%. Hyperpolarized [15N3]metronidazole can be potentially employed for hypoxia sensing. We also demonstrate P13C = 14.3% in [1-13C]pyruvate, a potent contrast agent for probing elevated anaerobic glycolysis.

POSTER 080
Order-unity 13C Hyperpolarization of a-ketocarboxylates in Under 1 minute via SABRESHEATH: [1-13C]pyruvate, [1-13C]alpha-ketoglutarate and [1-13C]ketoisocaproate
Presenting Author: Isaiah Adelabu

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NMR spectroscopy and imaging have become clinical tools for non-destructive diagnostic imaging of diseases. NMR hyperpolarization enhances sensitivity of MRI by 4-6 orders of magnitude, therefore, enabling real-time metabolic imaging. Hyperpolarized [1-13C]pyruvate and other [1-13C]alpha-ketocarboxylates are the key molecular probes for imaging of central biochemical pathways frequency upregulated in cancers and other diseases. Here we report on the utility of Signal Amplification by Reversible Exchange in SHield Enables Alignment Transfer to Heteronuclei (SABRE-SHEATH) as low-cost ($20k) and rapid (1 minute) hyperpolarization technique for efficient production of hyperpolarized [1-13C]pyruvate, [1-13C]alpha-ketoglutarate and [1-13C]ketoisocaproate with 13C polarization of up to 18%. We anticipate that the use of pulsed-SABRE-SHEATH and SABRE catalyst purification technologies will enable production of highly polarized biocompatible solutions.

POSTER 081
Protein NMR by dissolution DNP approaching physiological concentrations - Revealing dilution-induced conformational changes.
Presenting Author: Dennis Kurzbach

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NMR is the only method that allows
Complete structural characterization with atomistic resolution in the native solution environment of most biomacromolecules. However, due to the intrinsically low sensitivity of the method, the target concentrations are much higher than under physiological conditions. Herein we demonstrate how to obtain NMR spectra of the the central transcription factor MAX (MYC-associated factor X) at a concentrations below 1 micromolar. To this end, we dissolved the protein in “hyperpolarized water”. This boosts the signal intensity by orders of magnitude enabling access to very low concentration regimes. The importance of this methodological advance is demonstrated by the fact that the MAX homodimer dissociates and adopts another monomeric conformation under physiological conditions compared to the high-concentration regime.

POSTER 082
Integrated, Stretched and Adiabatic Solid Effects
Presenting Author: Yifan Quan

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Here we present a theoretical description of DNP induced by an arbitrary frequency-swept microwave pulse. It shows that a strong microwave sweep can be highly efficient and transfer twice the electron polarization to the surrounding nuclei. The theory is used to explain the ISE, SSE and ASE and experimentally verified at 9.4 GHz (0.34 T), showing that SSE and ASE can be more effective than

the ISE. At high magnetic fields, where the EPR line width is narrower than the nuclear Larmor frequency, the theory predicts that SSE can be optimally efficient. In addition, we show that the physics underlying the ISE, SSE and ASE is similar and we provide improved definitions to distinguish the three mechanisms from one another.

POSTER 083
TinyPol-like Biradicals for Highly Efficient DNP MAS NMR at High Magnetic Fields and Fast MAS
Presenting Author: Georges Menzildjian

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In this presentation, we will report recent advances in the design of polarizing agents for efficient DNP MAS MNR at very high magnetic fields and fast spinning frequencies. Notably, new TinyPol binitroxides with finely-tuned geometry will be introduced and their DNP performance will be presented at 18.8 T. The best radical in this new series outperforms current binitroxides, with enhancement factors as high as 170 at 60 kHz MAS. These results are interpreted jointly with EPR data to disentangle the key structural parameters controlling the DNP cross-effect efficiency at high fields.

POSTER 084
RASER MRI: Magnetic Resonance Imaging with Nonlinearly coupled Point Spread Functions
Presenting Author: Sören Lehmkuhl

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The spatial resolution of MRI is fundamentally limited by the width of Lorentzian point spread functions (PSF) associated with the exponential decay rate (1/T2*). Here we overcome the traditional T2* limit by establishing RASER (Radio-frequency Amplified Stimulated Emission of Radiation) in imaged media. RASER imaging bursts emerge when placing spins with sufficient population inversion a weak magnetic field gradient without applying RF pulses. Such bursts develop through cooperative nonlinear interaction, which can resolve structures of the imaged object at a higher spatial resolution. The nonlinear physics of RASER MRI comprises artifacts such as line collapse due to mode locking, side lobes outside of the imaging domain and amplitude distortions.

POSTER 085
Understanding and Optimizing Dynamics in SABRE Hyperpolarization
Presenting Author: Jacob Lindale

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Signal Amplification By Reversible Exchange, or SABRE, is a rapidly growing hyperpolarization method that converts the singlet order of parahydrogen, p-H2, into observable spin order (magnetization or multi-spin order) on a target during reversible interactions with an iridium catalyst. It became clear that the exchange processes would have to be considered in the design of next generation SABRE experiments. To address this, we
Completely derived the exact Dissipative Master Equation (DMEx) for chemical exchange, which offered a significantly improved convergence radius over the traditional master equation at no additional computational cost. Coupling computational optimization to experimental and conventional theoretical methods helped us unravel the underlying dynamics and highlight new avenues towards both optimizing and extending the scope of SABRE.

POSTER 086
SABRE Enhancement with Oscillating Pulse Sequences: Symmetry Reduces Robustness
Presenting Author: Xiaoqing Li

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SABRE (Signal Amplification by Reversible Exchange) methods provide a simple, fast, and cost-effective method to hyperpolarize a wide variety of molecules in solution. Here, we present several oscillating pulse sequences that use magnetic fields far away from the resonance condition of continuous excitation. An analysis with average Hamiltonian theory indicates that the oscillating pulse, in effect, adjusts the J-couplings between hydrides and target nuclei and that a much weaker coupling produces maximum polarization. This theoretical treatment, combined with simulations and experiment, show substantial magnetization improvements relative to traditional X-SABRE methods. It also shows that, in contrast to most pulse sequence applications, waveforms with reduced time symmetry in the toggling frame make magnetization generation more robust to experimental imperfections.

POSTER 087
Proton-only detection of hyperpolarized 13C2-pyruvate by adiabatic SLIC pulses
Presenting Author: Iuliia Mandzhieva

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Hyperpolarized magnetic resonance imaging (HP MRI) has enabled in vivo imaging of biologically active molecules during metabolic processes. To date, Pyruvate is the principal HP substrate because it plays a central role in vital metabolic pathways and could be used as biomarker for various diseases. However, HP MRI detection requires specialized 13C capabilities.
Here we present a method to make HP MRI compatible with scanners that only have proton capabilities. SABRE-SHEATH creates HP carbon singlet on 1-2-[13C2]-Pyruvate. Then, Spin-Lock Induced Crossing (SLIC) pulse transfers HP from 13C2 singlet to the methyl group protons.
This method paves the way for HP 1H MRI detection for existing MRI scanners and future widely used low-field MRI machines establishing an affordable molecular imaging platform.

POSTER 088
Hyperpolarized 29Si Isotope-Enriched Silicon and Silica nanoparticles for Development of High Resolution 29Si MRI imaging probe
Presenting Author: Jiwon Kim

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Silicon and silica nanoparticles have garnered attention as promising biomedical probes for hyperpolarized 29Si magnetic resonance imaging and spectroscopy. In this work, the development of MR imaging probe based on 29Si isotope-enriched silicon and silica nanoparticles using DNP-NMR is discussed. The result exhibits that the higher 29Si ratio, the more effective signal amplification was observed, demonstrating its applicability for in vivo MR imaging. Furthermore, we demonstrated that the polarization penetration depth of core-shell structured SiO2 NPs corresponds to ~10 nm, and the higher signal intensity appear as 29Si ratio in the shell increases. This study is expected to serve as important basic research for building a therapeutic system based on silicon/silica nanoparticles using DNP in the future.

POSTER 089
Spatiotemporal Denoising to Improve Image Quality in Large Coverage Abdominal Hyperpolarized [1-13C]Pyruvate Studies
Presenting Author: Tanner Nickles

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A major limitation in hyperpolarized (HP) ^{13}C MRI is the finite T_1 relaxation time and signal-to-noise ratio (SNR) of downstream metabolites, which restricts the achievable spatial resolution. To overcome this limitation in abdominal studies, a patch-based spatiotemporal denoising method has been successfully applied to denoise dynamic imaging data in [1-^{13}C]pyruvate echo-planar imaging (EPI) human datasets. With denoising, a 10+ fold sensitivity gain was achieved for all metabolites in the HP [1-^{13}C]pyruvate datasets, which was qualitatively most apparent for [1-^{13}C]alanine and late-phase [1-^{13}C]lactate. With the increase of SNR of HP [1-^{13}C]pyruvate human abdomen datasets, spatiotemporal denoising also demonstrated improvements in quantification of downstream metabolite conversion-rates.

POSTER 090
Some Design Parameters for DNP Polarizing Agents
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Although the hyperpolarization of solids is now routinely achieved by dynamic nuclear polarization (DNP), DNP enhancements obtained with standard biradical polarizing agents are still much lower than the theoretical limit. Here we report a series of deuterated TEKPol biradicals to understand the effect of selective deuteration on the transfer of hyperpolarization into the bulk. Increasing deuteration leads to decreased DNP enhancements, underscoring the key role of protons in the biradical molecule in the spin diffusion process.
As a promising alternative to nitroxide-based radicals, we have also synthesized a series of Gd(III) complexes to validate the inverse square dependence of the DNP enhancements on the Gd(III) zero-field splittings, paving the way for the development of improved polarizing agents.

POSTER 091
Unexpected Inversion of Hyperpolarized ^{13}C NMR Signals Through Cross-Correlated Cross Relaxation in Dissolution DNP Experiments
Presenting Author: Mattia Negroni

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During the exploration of new possibilities for the use of Dissolution Dynamic Nuclear Polarization (DDNP) we stumbled across an unusual phenomenon: after polarization and subsequent dissolution, protonated hydrocarbon moieties display inverted ^{13}C signals compared to ^{1}H while deuterated species do not. The explanation is based on cross-correlated cross relaxation (CCR) coupled to very high proton polarizations that induce a selective inversion due to low field passages during the sample transfer. Parallel detection of ^{1}H and ^{13}C allows the monitoring of the signals evolution and the results were confirmed by relaxation simulations. The ubiquitous presence of CH spin systems made this phenomenon quite common and might be exploited for selective spectroscopic labeling.

POSTER 092
Purified Parahydrogen-Hyperpolarized Pyruvate and Fumarate for Preclinical in-vivo Metabolic Magnetic Resonance Imaging
Presenting Author: John Blanchard

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Metabolic MRI using hyperpolarized 13C nuclei is becoming a popular tool for the study of metabolic processes using MRI. In recent years ParaHydrogen Induced Polarization (PHIP) has been explored as a fast, cost-efficient method for the preparation of hyperpolarized metabolites. Here we present an in vivo study in tumor-bearing mice models using fumarate and pyruvate polarized via PHIP and subsequently purified, delivering clinically relevant concentrations (>100mM) with polarization values up to 20 % at the time of injection. Using a fully automated NVision polarizer/purification system we demonstrate that PHIP can compete with dissolution Dynamic Nuclear Polarization (dDNP). The presented advances allow for affordable and scalable hyperpolarized preclinical MRI and the development of a clinical PHIP based hyperpolarization platform is underway.

POSTER 093
Spin dynamics of [13C2]pyruvate hyperpolarization by parahydrogen in reversible exchange at uT fields
Presenting Author: Austin Browning

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Hyperpolarization of pyruvate via Signal Amplificaton By Reversibble Exchange (SABRE) is currently receiving a lot of attention because of the relative ease of hyperpolarization and the central biological relevance of pyruvate as a biomolecular probe for in vitro or in vivo studies. Here the [1,2-13C2]pyruvate SABRE spin system and its field dependence are analyzed through analytical first principal analysis of the governing 4-spin dihydride-13C2 Hamiltonian, followed by spin dynamics simulations of the 7-spin dihydride dihydride-13C2-CH3 system, and comparison to experiment. These methods help unravel the observed spin state mixing of singlet states, triplet states and their combination with CH3 magnetization states to arrive at a greater understanding of the [1,2-13C2]pyruvate-SABRE system and how to optimally control it.

POSTER 094
SABRE Optimization Leads to Over 10% Hyperpolarization on Current Generation Cancer Drugs
Presenting Author: Keilian MacCulloch

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DNP hyperpolarization is unlocking doors to study complex biophysics of protein interactions using NMR. The increased sensitivity in selected nuclei afforded by hyperpolarization allows characterization of interactions that are otherwise not observable. SABRE is a cheap and quick hyperpolarization alternative to DNP, however it generates lower levels of hyperpolarization on a limited substrate scope. SABRE efficiency is controlled by exchange rates, J-couplings, and Larmor frequencies of the spin system. Thus, distinct substrates will have a unique optimal parameter set. Fine tuning these parameters for individual substrates is instrumental for efficient SABRE hyperpolarization. Here, we extended SABRE hyperpolarization to 15N nuclei on cancer drugs. Optimization of the polarization process generated >10% hyperpolarization and T1 lifetimes over seven minutes for both drugs.

POSTER 095
Continuous Parahydrogen Pumped RASER Enabled by Hydrogenations in a Semipermeable Membrane Flow Reactor
Presenting Author: Christopher Nelson

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In 2017, the first parahydrogen pumped RASER (pH2-RASER) was described using signal amplification by reversible exchange (SABRE). Here we report on pH2-RASER induced by hydrogenation reactions, expanding beyond the reversible exchange of SABRE. Since hydrogenation is not reversible, a flow reactor setup was created to allow for a continual replenishing of the population inversion responsible for the RASER effects, while addressing shortfalls in previous studies which paused acquisition in order to bubble pH2 gas.

We have collected 65.5s of continuous RASER signal with a ~1T (43.26Hz) magnet and achieved line widths of 0.019Hz, which are within 1% of the theoretical limit, given the acquisition time and the strength of the magnetic field.

POSTER 096
Parahydrogen Induced Polarization for Precision Measurements and Molecular Imaging
Presenting Author: Thomas Theis

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Parahydrogen induced polarization (PHIP) is cherished for its relative simplicity and ease. Here we report on the most recent progress with PHIP in hydrogenative addition and non-hydrogenative reversible exchange reactions to attain (a) Radiofrequency Amplification By Stimulated Emission of Radiation (RASER) for precision measurements of J-couplings and chemical shifts and (b) molecular imaging with a cryogen-free, variable field MRI system after SABRE-SHEATH hyperpolarization of 13C-pyruvate to above 10% polarization with a temperature cycling approach. Simultaneously, we continue the substrate scope expansion to other metabolites and drugs including antifungals and cancer drugs.

POSTER 097
Characterization of the Microwave Field Distributions within Photonic Band-Gap Resonators by DNP NMR of Thin-Film Samples
Presenting Author: Alex I. Smirnov

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One-dimensional photonic band gap (PBG) resonators are effective in focusing B1e fields over flat thin film samples. We demonstrate up to 50-fold enhancement in microwave power over thin film samples of ca. 5 mm in diameter by measuring the DNP enhancements at 7 T magnetic field for natural 13C abundance of microdiamonds embedded in commercial 3-4 mil lapping films. We have investigated B1e field distribution inside a PBG resonator made of alternating sapphire and quartz plates. COMSOL simulations guide optimization of the sample configurations for the optimal signal intensity. Up to 2.2-fold greater DNP-enhanced NMR signal was achieved by placing two films on the last sapphire plate and one film on the mirror as compared to a single-layer sample.

POSTER 099
An Enhanced Overhauser Effect following Selective Deuteration of BDPA
Presenting Author: Michael Mardini

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Our previous findings support the notion that the varying hyperfine couplings of the protons on BDPA result in different contributions to the overall observed Overhauser effect enhancement. Here, we use selective deuteration to remove the weakly coupled phenyl protons which contribute negative enhancement. DNP NMR experiments on the selectively deuterated derivative yield an OE enhancement nearly double in magnitude relative to the fully protonated radical. These results further corroborate that the OE is indeed the active mechanism, and provide an avenue for improved sensitivity when using BDPA or its derivatives for DNP.

POSTER 100
Echo-planar Imaging of pH In Vivo Using Hyperpolarized [13C]Bicarbonate
Presenting Author: Yaewon Kim

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Acid-base imbalance in body fluids can indicate abnormal cellular processes caused by various pathological states such as inflammation and cancer. Thus, a reliable tool for non-invasive pH measurement is an unmet clinical need. In this study, we validated a pH imaging method utilizing hyperpolarized [13C]bicarbonate (HCO3-) and CO2 in phantoms and then applied this technique to measure pH of the rat kidney on a 3T clinical MRI. By employing a metabolite-selective EPI sequence with multiple acquisitions and a modified Henderson-Hasselbalch equation, pH maps were obtained from the summed images of HCO3- and CO2. This resulted in a >3-fold SNR gain compared to a single-scan measurement, supporting data acquisition at higher spatial resolution to facilitate accurate determination of local pH changes.

POSTER 101
Observation of Anomalously Large Anti-Phase Signals from Hyperpolarized Orthohydrogen Using a Heterogeneous MOF-Based SABRE Catalyst
Presenting Author: Drew Brittin

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Hyperpolarized orthohydrogen (o-H2) is a frequent byproduct of parahydrogen-based hyperpolarization approaches like SABRE, where the hyperpolarized o-H2 signal is usually absorptive. We describe a novel manifestation of this effect wherein large antiphase o-H2 signals are observed, with 1H enhancements up to ~600-fold (PH~2%). This anomalous effect is attained only when using an intact heterogeneous catalyst constructed using a metal-organic framework (MOF), and is qualitatively independent of substrate. This seemingly paradoxical observation is analogous to the “partial negative line” (PNL) effect recently explained in the context of PHIP by Ivanov and co-workers. The two-spin order of the o-H2 resonance is manifested by a two-fold higher Rabi frequency, and the lifetime of the antiphase HP o-H2 resonance is extended by nearly 3-fold.

POSTER 102
Time-resolved 129Xe Thermometry for Monitoring RF Heating
Presenting Author: David Hernandez

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Intensive application of RF pulses such as in saturation transfer techniques can cause unwanted heating that requires sensitive non-invasive tools to monitor this effect. The chemical shift of hyperpolarized Xe provides high NMR sensitivity even in aqueous solutions. We introduce a hardware setup using 129Xe NMR and optical thermometry that monitors heating for cw RF irradiation for various duty cycles with high sensitivity (e.g., 50 mK temperature change). It will be used in the future to compare RF heating of CEST implementations.

POSTER 103
Non-Intuitive Pulse Sequences Enhance SABRE Singlet Pumping
Presenting Author: Shannon Eriksson

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Diazirine functional groups could make for promising NMR-based biological molecular tags that can support a long-lived nuclear state. Direct pumping of 15N singlet states was previously demonstrated in continuous-wave SABRE-SHEATH experiments, but is limited by an inability to directly couple a magnetic field to the singlet state. Here we show that, despite this limitation, a symmetric square pulse sequence with a 0 μT average generates significant improvements over CW-SABRE. Under Average Hamiltonian Theory, this oscillating field adds new phase-rotated J_NH terms to the high-field CW-SABRE Hamiltonian. By adjusting the pulse sequence parameters, we can further optimize the singlet enhancement pathways without the need to couple to the singlet state, yielding up to 50% enhancement beyond CW-SABRE experiments.

POSTER 104
Improving Parahydrogen Conversion Through Boiling Point Suppression of Liquid Nitrogen
Presenting Author: Nicholas Whiting

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The use of parahydrogen (pH2) to enhance NMR and MRI signals for a diversity of samples has recently increased in popularity. While liquid nitrogen-cooled pH2 generators are attractive due to their low cost of entry, there are benefits to having access to >50% pH2 for MR applications. Here, we introduce two approaches to increase pH2 fractions through lowering the liquid nitrogen temperature. The first approach used vacuum-mediated boiling point suppression, which achieved liquid nitrogen temperatures at its triple point (63 K) and produced ~65% pH2. The second approach used internal evaporation into injected helium bubbles to cool the liquid nitrogen to ~70 K, resulting in ~59% pH2. Both approaches are relatively low-cost modifications to existing liquid nitrogen-cooled pH2 generators.

POSTER 105
Reconversion of Parahydrogen Gas in Surfactant-Coated Glass NMR Tubes
Presenting Author: Robert Chimenti

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Once generated, parahydrogen gas (pH2) will reconvert to its normal isomeric abundance over time. While this may take weeks when stored in an aluminum cylinder, the reconversion is typically within hours inside glass NMR tubes; this is due to paramagnetic impurities within the glass itself. Here, we explore the role of surfactants that can coat the inside of NMR tubes and serve as a barrier between the pH2 and paramagnetic impurities. We used in situ Raman spectroscopy to monitor pH2 reconversion in NMR tubes coated with various silane-based surfactants and found that most extended the pH2 reconversion time by 1.5-2x. This may be of interest to researchers looking to extend the lifetime of pH2 in the gas phase.

POSTER 106
Two-Orders-of-Magnitude Improvement in SEOP Production (P*N) of Hyperpolarized 131Xe and Measurement of 131Xe and 129Xe Spin-Dependent Neutron Scattering Lengths via Pseudomagnetic Precession
Presenting Author: Abdulbasit Tobi Gafar

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We report on SEOP hyperpolarization of 131Xe and its potential for use in targets for neutron beam experiments. In ~100 cc Pyrex cells, 131Xe polarization up to 7.6±1.5% was achieved in ~8.5´1020 spins, corresponding to >100-fold improvement in (P*N) compared to previous efforts. For neutron science experiments we prepared larger cells (~300 cc) made of aluminosilicate glass. Calibrated using a 3He cell, P(129Xe)=17.6±0.8% and P(131Xe)=1.9±0.15% values were determined and used to perform the first measurements of the 131Xe and 129Xe spin-dependent neutron scattering lengths via pseudomagnetic precession. These values will help determine the sensitivity of envisioned Time Reversal Invariance Violation experiments on the 3.2 eV p-wave resonance of 131Xe in the search for new physics beyond the Standard Model.

POSTER 107
Optimization of α-Ketoglutarate Hyperpolarization with SABRE Elucidates Ideal Parameter Sets for α-Keto Acids
Presenting Author: Stephen J. McBride

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In this work, we demonstrate the SABRE hyperpolarization of a new α-keto acid substrate, α-ketoglutarate (AKG). We optimize the 13C polarization of AKG above 5% in methanol by probing the pressure, concentration, and temperature dependencies and examine the relaxation and polarization buildup dynamics. The optimization, relaxation, and buildup studies conducted shed new light on the chemistry and exchange dynamics of these α-keto acid SABRE systems, specifically highlighting how modulation of exchange rates in the low-temperature regime can enable very high levels of bound substrate polarization that drives free substrate polarization once the temperature is raised. Finally, to probe the applicability of AKG in in vivo MRI studies, we explored the SABRE dynamics of AKG in different ethanol/water mixtures.

POSTER 108
Exploring SABRE-Enhanced Imaging with a Portable Clinical Low-field MRI Scanner
Presenting Author: Nadiya Iqbal

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Most clinical MRI scanners have expensive, bulky, immobile, and confining magnets. Low-field (LF)MRI can obviate these limitations, but traditionally suffers from low sensitivity. The sensitivity boost provided by hyperpolarization is highly synergistic with LFMRI. We are investigating the potential for adapting a portable, point-of-care 64 mT clinical MRI scanner with SABRE hyperpolarization using a continuous pH2-blubbling setup. 1H SABRE detection of different samples (e.g. pyrazine) via LFMRI is compared with a benchtop NMR spectrometer. A few-hundred fold MRI enhancement is estimated by comparing the signal to that of pure water. We are currently working to confirm the origin of the HP signals, optimize sequences for rapid SABRE imaging, and enable continuous delivery of HP solutions created using ex situ SABRE.

POSTER 109
Scalar Relaxation in 13C Dynamic Nuclear Polarization in Liquids
Presenting Author: Tomas Orlando

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Dynamic nuclear polarization (DNP) in liquids can increase ^{13}C-NMR signals more than 100-fold at high magnetic fields (≥ 3.4 T), thanks to the large electron-nuclear scalar relaxation. Nevertheless, the lack of a reliable strategy to predict scalar enhancements conveyed the idea that scalar relaxation occurs only in exceptional cases.
Here we present three different modeling strategies for scalar relaxation, which are based on nuclear relaxation theory and molecular dynamics. We analyzed the case of CHCl3 doped with nitroxide radical, and identify correlation times for collisional processes in the range τ ~ 0.5-30 ps. Besides random molecular collision, transient interactions based on hydrogen bonds can be effective in driving scalar relaxation and thus large enhancements in liquids.

POSTER 110
Double resonance 13C-1H Overhauser DNP at 14T
Presenting Author: Thierry Dubroca

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DNP in liquids is challenging, particularly at high magnetic fields, where resolution is needed to solve scientific questions. We present here a large sample volume double resonance (13C-1H) liquid DNP probe capable of delivering significant microwave power at the sample. This newly developed probe is applied to scalar Overhauser DNP, that polarize mostly the nuclei of heavier, electron-rich atoms. This leaves 1H NMR outside the realm of direct application. This work shows an experiment using 13CHCl3 comixed with TEMPO that can deliver 1H-detected NMR experiments, such as INEPT and two-dimensional HSQC, via scalar Overhauser DNP enhancements from 13C. In general, extensions of this approach might prove advantageous for enhancing even further the sensitivity provided by Overhauser DNP.

POSTER 111
Evaluating Absorption Spectroscopy for Measuring Rubidium Vapour Densities in a 129Xe-Rb Polariser
Presenting Author: James Ball

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The Rubidium vapour density, [Rb], within the optical cell is a key parameter affecting 129Xe-Rb SEOP polariser performance and has been measured to be lower than saturation levels within some 129Xe-Rb polarisers. This work assesses the use of absorption spectroscopy to measure [Rb] in a clinical-scale 129Xe-Rb polariser. Violet transitions (52S1/2 to 62P1/2 and 62P3/2) and near-IR transitions (52S1/2 to 52P1/2 (D1) and 52P3/2 (D2)) were probed. Results show that under high [Rb] conditions in our setup, specifically near the Rb pool source, [Rb] determined from the highly attenuating D1 and D2 absorption lines are underestimated when compared to [Rb] calculated from the less attenuating violet transitions. This suggests a breakdown in Beer's law when probing the near-IR transitions.

POSTER 112
Intermetallic Nanoparticle Catalysts Afford >20% Pairwise Selectivity in Parahydrogen Based Hyperpolarization
Presenting Author: Tommy Zhao

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Supported metal nanoparticle catalysts offer key advantages over homogeneous catalysis for applications of parahydrogen-based hyperpolarization. The facile separation from the solid catalyst affords continuous production of contaminant-free hyperpolarized biomolecules. However, signal enhancements achieved by monometallic nanoparticles have been modest due to the stepwise addition mechanism and fast H ad-atom diffusion. Recent work in our lab has demonstrated that the pairwise selectivity can be significantly increased using certain binary intermetallic phases composed of an active (Pt, Rh) and an inert (Sn, In) metal. Superior pairwise selectivity and NMR signal enhancements are obtained using PtSn@mSiO2 and RhIn3/SBA-15 intermetallic nanoparticle catalysts. These catalysts yield unprecedented pairwise selectivities of 19.7% in propyne hydrogenation and 20.7% in propene hydrogenation, respectively.

POSTER 113
Towards Biocompatible SABRE Solutions by Optimizing a Ir-NHC Water Soluble Catalyst
Presenting Author: Mustapha Abdulmojeed

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Signal Amplification by Reversible Exchange (SABRE) is a cost effective, fast and reversible technique to hyperpolarize relevant biomarkers for MR Imaging. However clinical applications of this method have been hindered due to solvent and catalyst toxicity. New water soluble catalysts that enable direct SABRE hyperpolarization in aqueous media offers a viable pathway to in vivo SABRE experiments. Here, an attempt is made to optimize SABRE hyperpolarization in D2O, by electronically modifying the imidazole core of a water soluble Ir-NHC catalyst, through the synthesis of a derivative with doubly methylated imidazole backbones. These two catalysts

were further optimized for catalyst: substrate (Nicotinamide) ratio, polarization transfer field (PTF) and solvent effect. Preliminary data show optimum enhancement with 10:1 substrate to catalyst ratio.

POSTER 114
Versatile Liquid Shuttling System Incorporating an Ultrasonic Spray Injection Reactor for ALTADENA-Type Hyperpolarization Experiments
Presenting Author: Maria-Jose Ferrer

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We demonstrate an ultrasonic spray injection reactor system interfaced with a liquid shuttling system incorporating a dual syringe pump and a flow NMR probe. This system was used to acquire 1H and 13C spectra resulting from side-arm hydrogenation (SAH) of propargyl pyruvate and propargyl acetate with 99% parahydrogen. Spin order transfer to 13C of pyruvate and acetate was achieved using our level anti-crossing (LAC) ALTADENA INEPT hybrid technique. The efficiency is compared to other spin order transfer schemes

POSTER 115
PHIPNOESYS: A System for Intermolecular Nuclear-Overhauser-Effect-Mediated Transfer of Parahydrogen-Induced Polarization
Presenting Author: John W. Blanchard

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The most promising path to increased NMR sensitivity seems to involve moving beyond equilibrium polarization with so-called hyperpolarization techniques. We recently reported a new strategy for polarization enhancement where organic crystals are optically polarized and then dissolved in target solutions, allowing for transfer of hyperpolarization via the intermolecular nuclear Overhauser effect -- a system we call HYPNOESYS. Advances in parahydrogen-induced polarization (PHIP) have since led to our development of a PHIP-based HYPNOESYS -- or PHIPNOESYS – prototype. The system reacts an unsaturated precursor with parahydrogen, then converts the singlet order into in-phase 1H magnetization via a B0- or B1-field sweep. This hyperpolarized source material is then mixed with a solution of interest and transported into an NMR spectrometer for measurement.

POSTER 116
Mapping Hydration-layer on Surface of a Signaling Protein: Measuring “Dynamic Resonance” with ODNP
Presenting Author: Farhana Syed

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I am uploading a long abstract below.

POSTER 117
A field stability in cryogen-free magnets for solid state MAS NMR applications
Presenting Author: Jeremy Good

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Eugeny Kryukov (Cryogenic Ltd): Jeremy Good (Cryogenic Ltd.)

The European Regulatory Authority has declared a stop to the use of a natural gas in the near future. No natural gas means that no helium gas will be available and liquid helium cooled magnets will have to be replaced by cryogen-free cooled versions. The development of cryogen-free magnet technology for NMR applications is needed urgently. We show that a temporal field fluctuation

generated by a cryogen-free cryocooler can be reduced to less than one part per billion. In addition, the field settling time in such magnets can be brought to acceptable levels within an hour after the field ramp. This opens the possibility of using a single superconducting magnet at different fixed fields without compromising its NMR performance.

POSTER 118
Towards a Handheld Nuclear Quadrupole Resonance (NQR) System
Presenting Author: Peyman Dehghanzadeh

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Evaluating agricultural and pharmaceutical products for quality assurance is a significant concern for different organizations and agencies, such as the Food and Drug Administration (FDA). Spectroscopic approaches are preferred for authentication due to their specificity and potential for non-destructive analysis. Several techniques, including near-infrared spectroscopy (NIR), mid-infrared spectroscopy (Mid-IR), Raman spectroscopy, and NMR are commonly used for this purpose. However, these methods can be challenging for solid samples such as dietary supplements and vitamins: the optical methods suffer from sensitivity to coatings and packaging, while solid-state NMR requires expensive high-field instrumentation. Nuclear Quadrupole Resonance (NQR) spectroscopy is a promising alternative method for characterizing such solid samples.

POSTER 119
Hydrogel assemblies for segregated chemical reaction monitoring by microfluidic NMR
Presenting Author: Neil MacKinnon

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Compartmentalized chemical reactions enable high levels of process sophistication. A challenge is tracking the molecular content in situ and over time so that reaction dynamics can be understood. As a step towards addressing this challenge, we have developed an NMR-compatible microfluidic system that includes electrochemical functionality. This system was used to assemble a multilayered hydrogel capable of multiple biocatalytic reactions, enabling successful simultaneous monitoring of urea and glucose conversion as a function of time. At the microfluidic level, it was possible to extract enzyme kinetics starting from 3 and 0.3 micromoles of initial material, with a single 1H NMR spectrum requiring 9 min of acquisition time. This concept can be extended to cascaded reaction monitoring and NMR-coupled electrochemistry.

POSTER 120
Study of Different Dielectric Resonators Permittivities on SNR enhancement for MRI application
Presenting Author: Qing Yang

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Dielectric resonators with Ultra-High Dielectric Constant (uHDC) materials in different shapes were applied to enhance RF coils transmit efficiency and receive sensitivity at intrinsic resonance modes that have been used for RF transmission and reception. The resonance modes depend on different parameters such as HDC's permittivity. The effect of different permitivities of uHDC discs were studied through computer simulations. We showed, to achieve the best focusing effect into phantom, the design with uHDC material should operate around the first TE01δ mode and lower than the second mode with the highest permittivity. This research investigated the question as whether the uHDC materials can be used at the resonance condition along the conventional RF coils in comparison to the non-resonance cases of the same materials.

POSTER 121

1H NMR signals enhancement at room temperature using a homebuilt X-band Overhauser DNP setup
Presenting Author: Wirya Feizi

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Overhauser effect (O.E.) DNP involves transferring the high spin configuration of the electron spins to the nuclear spins by microwave radiation at the electron paramagnetic resonance (EPR) frequency of the free electron source. Here, we present the preliminary results of an instrument setup that uses an X-band OE DNP polarizer for enhancing the 1H nuclear magnetic resonance (NMR) signal of water doped with free radicals by tens to hundreds of times compared to the thermal NMR signal of the sample. Researchers used a variety of free radicals with varying EPR linewidths in this study. The ability of this technology to enhance NMR signals could improve the sensitivity of magnetic resonance imaging (MRI) signals for improved diagnosis.

POSTER 122
250 GHz / 380 MHz EPR / DNP / ENDOR Resonance Structure
Presenting Author: Ravi Shankar Palani

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DNP at high-fields would benefit from pulsed-DNP methods instead of continuous-wave DNP that is widely used today. A resonance structure capable of confining the high-frequency microwaves with a high Q-factor is necessary to realize shorter electron pulse-widths enabling pulse-DNP experiments. However, these are challenging to make as the cylindrical cavities with helical grooves, an ideal resonator for this application, scale in dimension with the wavelength and therefore are difficult to fabricate for applications beyond 200 GHz. This work discusses the design, fabrication, and performance of a TE011 resonator at 250 GHz that also serves as an RF coil for NMR detection.

POSTER 123
Ball-Shift Automation to Achieve Reproducible Mapping of Transceiver Coils
Presenting Author: Jose Luis Uribe

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Achieving homogenous radio frequency (rf) magnetic fields in solid-state NMR transceiver coils is crucial for maximizing sensitivity during experimentation. Conventionally, the manual ball-shift assay, measuring tuning frequency perturbations, has been used to map the rf coil profile. However, this manual apparatus makes it difficult to achieve adequate precision to ensure uniform steps, thus, reproducibility is hard to attain. To improve on this, I present an automated method that uses inexpensive and open-source equipment to create a modular, yet specialized tool. An Arduino UNO, in combination with other programmable hardware, are used to automatically perform this method, along with an automatized data collection scheme for hands-free operation.

POSTER 124
A Current-Mode Class-D Low-Voltage Power Amplifier for Portable NMR Systems
Presenting Author: David Ariando

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Miniaturized NMR relaxometry is of growing interest for portable applications. Receiver circuits, NMR controller, and magnet designs have been miniaturized to some extent. However, transmitter design hasn't seen as much progress in miniaturization. We propose a current-mode PA, that greatly reduce supply voltage requirements while maintaining high-bandwidth operation of a typical voltage-mode PA. The proposed architecture uses a GaNFET-based H-bridge circuit with a large inductor that acts as a current source for the circuit. The transmitter utilizes RF excitation dead-time to build up a higher excitation energy without a HV supply. The filter design ensures rapid pulse rise/fall times on the RF coil. The design is suitable for battery operation while delivering a high power output into the RF coil.

POSTER 125
Interleaved NQR Detection of Explosives using Atomic Magnetometers
Presenting Author: Darwin Quiroz

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Nuclear Quadrupole Resonance (NQR) spectroscopy, aka zero-field NMR, is useful for contraband detection. Serial detection of different materials, however, can become prohibitively lengthy, particularly when T1-times are long; for instance, T1=17 seconds for ammonium nitrate. Sensitive coil detection requires tuned probes with variation of multiple electrical components with each new frequency. With NQR frequencies that widely vary, rapidity and scalability become daunting. In contrast, an atomic magnetometer is tuned with a small magnetic field, making rapid adjustments possible. Using this advantage, atomic magnetometers were used for interleaved detection – where potassium chlorate is measured during the recovery time of ammonium nitrate. A double-resonance-excitation probe was used here, but could be replaced by broad-band excitation, since high-Q is unnecessary for detection.

POSTER 126
Potential of Laser Assisted Deposition of Copper for "3D Printing" NMR Microcoils
Presenting Author: Peter M. Costa

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NMR microcoils have greater mass-sensitivity in comparison to standard commercially available coils, allowing for the analysis of mass-limited samples. Current microcoil production methods such as electroplating copper are limited due to long and expensive processes. A potentially versatile technique for development of effective microcoils is laser assisted deposition of copper to deposit copper onto various substrates. This has the advantage where the laser can be utilized to directly "print" coils on inside or outside of transparent materials (i.e. NMR tubes) thereby increasing its filling factor. The results explore the potential of laser assisted copper deposition to develop a fully functional, and optimized microcoil for analysis of mass-limited samples.

POSTER 127
The Network for Advanced NMR: Democratizing NMR Spectroscopy
Presenting Author: Alexander L. Paterson

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The mission of NAN, the Network for Advanced NMR, is to provide state-of-the-art NMR instrumentation to investigators in the US and abroad to advance relevant areas of science. The NAN member institutions are acquiring two 1.1 GHz NMR spectrometers: one dedicated to the study of solids at the University of Wisconsin-Madison; and one dedicated to the study of solutions at the University of Georgia. A comprehensive open-source NMR Knowledgebase of protocols, pulse sequences, and example datasets is in development and will be shared through the NAN portal. The NAN portal is constructed by the University of Connecticut to facilitate access to high-field NMR instrumentation, expertise and data, and to advance the collective efforts of the NMR community.

POSTER 128
Localized shim system for improving NMR spectral resolution
Presenting Author: Yen-Tse Cheng

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High homogeneity of the static field B0 inside the NMR magnet is a crucial prerequisite for detecting the spin dynamics of the target nuclei to differentiate the components in a small sample volume. Commercial MRI magnets are less demanding in terms of field homogeneity compared to NMR spectroscopy and therefore they are usually not feasible to discern the peaks that have small chemical

shifts. In this contribution, we create a compact localized shim system, enabling a higher NMR spectral resolution. Our approach is established by local field correction within the bore of a 1.05 T permanent magnet MRI. The proposed system can improve spectral linewidth from 40 Hz to 12 Hz.

POSTER 129
A Further Increase in Sensitivity for a 13C-Optimized 1.5 mm Superconducting NMR Probe
Presenting Author: William W. Brey

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Direct detection of 13C is valuable for systems where 1H spectroscopy yields inadequate resolution, but the relatively poor sensitivity of 13C is a problem. We previously developed a high sensitivity probe for 13C detection of mass limited samples based on superconducting resonators. With a matched Q factor of 1300, the probe had outstanding 13C sensitivity compared to probes using normal metal coils and has been used for many previously inaccessible samples. Here we report that we have increased the Q factor to 4300 and the sensitivity has increased by 33%. With the higher Q, it has been necessary to implement overcoupling and a passive Q-switching technique to maintain uniform sensitivity over the 200+ ppm chemical shift range of 13C.

POSTER 130
Reverse-Wound Solenoid for X-Nuclei Detected Solid-State NMR at High Magnetic Fields
Presenting Author: Wenping Mao

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As NMR magnetic fields increase to 1.2 GHz and above, it becomes more difficult to tune large-volume multi-turn solenoidal coils to detect nuclei in 13C…11B range while still maintaining good RF efficiency. Here we utilize reverse-wound (RW) solenoid, which is composed of two multi-turn solenoids that adjoin axially but connect in parallel electrically, in order to raise tuning limits for efficient direct-detected solid-state NMR. We compare detection in RW coil to that in conventional straight-wound solenoid in a Low-E 1.5 GHz 1HX 3.2 mm MAS probe. The results show that in addition to a higher tuning limit, the RW solenoid has a similar B1 homogeneity but lower electric field. Therefore, use of RW solenoid has sensitivity advantage for biological samples.

POSTER 131
Towards Improving Field Regulation in the 35.2 T Series-Connected Hybrid Magnet
Presenting Author: Jeffrey Louis Schiano

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Powered magnets provide high magnetic fields which present new opportunities for NMR spectroscopy. However, a field regulation system is required to reduce temporal field fluctuations induced by the power supply and cooling system. The present flux regulation system reduces field fluctuations to 0.3 ppm peak-to-peak. Modification of the feedback algorithm at the existing sample rate has not yielded additional reductions. Work is under way to improve field regulation by both increasing the sample rate and investigating mechanisms responsible for limiting performance. This work quantifies the field inhomogeneity of the correction coil and its effect on the field regulation system. We also compare the field estimate acquired using the primary and external lock channels of the probe.

POSTER 132
Sensitive Home-built 1.3 mm 1H/X/Y MAS Probe for 1H-Detection
Presenting Author: Peter L Gor'kov

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We report on the home-built 1.3 mm MAS probe constructed for 1H-detected applications in biological solids at 60+ kHz MAS speeds. The 800 MHz triple-resonance HXY probe uses home-made 1.3 mm spinning module designed around air bearings purchased from Bruker-Biospin. This presentation will describe optimization of 1H-detecting RF circuit that yields 2X higher signal-to-noise when compared to the commercial 1.3 mm Bruker probe we own. For the benefit of biological experiments, we also improved probe's ability to cool samples down to 0°C, as compared to +30°C limit in our commercial Bruker probes. We can also tune our probe to many isotopes in 109Ag...31P range, in triple-resonance HXY or in double-resonance HX configurations.

POSTER 133
Software Advances Allowing Smoother ODNP Data Acquisition
Presenting Author: Alexandria Guinness

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Overhauser Dynamic Nuclear Polarization (ODNP) has become a method widely used by groups studying protein conformations, hydration dynamics, and intricate confined systems, such as reverse micelles and porous materials. Yet, it is still an emerging technique undergoing vast improvements that enable data collection with greater flexibility and accuracy.
We have developed software that interfaces with a modular instrument and is capable of synchronizing NMR collection (with a SpinCore transceiver and several home-built components) with changes in microwave powers (controlled by a Bridge 12 microwave power source), B_0 fields (controlled by a commercial electromagnet), and eventually temperatures.

POSTER 134
A Cryogenically-Cooled High-Sensitivity Nuclear Quadrupole Resonance Spectrometer
Presenting Author: Soumyajit Mandal

Complete Author List:
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Over the last several years we have described our development efforts toward a high sensitivity nuclear quadrupole resonance (NQR) spectrometer, notably by i) reducing thermal noise ($4kTR\Delta f$) in the detector by cryogenic cooling in liquid nitrogen at 77 K and ii) building a digitally-tuned reconfigurable series/parallel-resonant probe which maximizes transmitter efficiency and reciever noise factor. Recently we have
Completed implementation of the spectrometer and shown that it achieves a signal-to-noise ratio (SNR) enhancement factor of 88.4×. Our
Complete cryogenic spectrometer also implements i) a custom class-D power amplifier with GaN FET output stage; ii) a custom low-noise broadband JFET-input pre-amplifier; and iii) a microcontroller for generating digital control signals.

POSTER 135
Application of a 1mm Micro-coil Flow Probe with External Lock for In-Vivo NMR Metabolomics of Daphnia Neonates and Dormant Eggs
Presenting Author: Monica Bastawrous

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In-Vivo NMR provides unprecedented information on biochemical changes within living organisms in response to external stressors. Recent work argues D. magna dormant eggs and neonates are especially sensitive to toxins, making them critical for the species long-term survival. Due to their sizes, ~350um O.D. eggs and ~1000um length neonates, analysis using traditional 5mm NMR probes is challenging. Here, a 1mm NMR probe, with external lock, is used to analyze dormant eggs and neonates using a novel uL volume flow system. High quality metabolic fingerprints and responses are extracted from the 2D HSQC of 13C enriched dormant eggs and neonates. The main presentation will show detailed metabolic profiles of living organisms and demonstrate the potential to detect and explain environmental stress.

POSTER 137
Identifying the Metabolic Signatures of PPARD-Overexpressing Gastric Tumors
Presenting Author: Shivanand Pudakalakatti

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Effective strategies for gastric cancer prevention and treatment, as well as understanding the drivers of this often-fatal disease, are critically needed. Peroxisome proliferator-activated receptor delta (PPARD) is a ligand-dependent nuclear transcription factor that regulates a multiplicity of pathophysiological processes. Recent discovery of PPARD-overexpressed in villin-positive gastric progenitor cells, demonstrated spontaneous development of large, invasive gastric tumors as the mice aged. These unique animal models allowed us to address the knowledge gap of PPARD-regulated downstream metabolic changes and determine the significance of changes in gastric tumorigenesis by hyperpolarized MR spectroscopy, NMR spectroscopy and LC-MS. Unlike many cancer systems, we found PPARD-overexpressing gastric cancer tumors are not primarily dependent on aerobic glycolysis but dependent on fatty acid oxidation as the bioenergy source.

POSTER 138
DREAMTIME NMR Spectroscopy: Targeted Multi-compound Selection with Improved Detection Limits
Presenting Author: Andre Simpson

Complete Author List:
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An NMR approach, termed DREAMTIME, is introduced that provides “a molecular window” inside complex systems, capable of showing only what the user desires, with
Complete molecular specificity. The user chooses a list of molecules of interest, and the approach detects only those targets while all other molecules are invisible. The approach is demonstrated in whole human blood/urine, small living aquatic organisms in 1D/2D NMR and MRI. Finally, as proof-of-concept, once overlap is removed via DREAMTIME, a novel “multi-focusing” approach can be used to increase sensitivity. In human blood and urine, sensitivity increases, 7-12 fold, over standard 1H NMR are observed. Applicable even to unknowns, DREAMTIME has widespread application, from monitoring product formation in organic chemistry to monitoring molecular targets in-vivo.

POSTER 139
Tracking the Effects of Using Deuterated Water in Cell Culture on the Glycolytic Pathway of Neuroblastoma and Glioblastoma Cells Using 13C NMR
Presenting Author: Cody Larsen

Complete Author List:
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Heavy water has been found to have a significant impact on cell proliferation and growth, with hyper-deuterated and deuterium depleted growth media demonstrating chronic cytotoxicity and a decreased proliferation rate for cells. This work seeks to investigate the effects that D2O has on the glycolytic pathways of cultured neuroblastoma and glioblastoma cells. Various concentrations of D2O were administered to cultured cells and the metabolic pathways were studied via 13C NMR spectroscopy through the conversion of 13C glucose to lactate over 48 hours using a Bruker 600 Hz NMR spectrometer.

POSTER 140
13C NMR Investigation of the Pentose Phosphate Pathway in Non-Small Cell Lung Cancer Cells
Presenting Author: John Michael B Tubije

Complete Author List:
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Normal cells produce energy mostly through mitochondrial oxidative phosphorylation (OXPHOS). Compared to normal cells, cancer cells exhibit high rate of glucose utilization under the glycolytic pathway for ATP production despite the presence of oxygen for OXPHOS. To further understand the aberrant metabolic behavior of cancer cells, we are conducting experiments to explore ribose as an alternative source, and the viability of the hexose monophosphate shunt as an alternative pathway for ATP production in cancer cells. Initial NMR spectroscopy results of A549 Non-Small Cell Lung Cancer (NSCLC) cells doped with 13C ribose indicate that 13C Ribose was metabolized. Results of this study will be presented.

POSTER 141
Effect of Hypoxia on Glycolysis in Cultured Colorectal Cancer Cells Studied by 13C NMR Spectroscopy
Presenting Author: Daniel Anable

Complete Author List:
Daniel Anable (University of Texas at Dallas); Lloyd Lumata (University of Texas at Dallas)

Hypoxia is a physiological condition that is detrimental to normal cells in which there is a lack of oxygen. Cancer cells however thrive even under hypoxic conditions. In this study, we have investigated the effects of total hypoxia (0% O2) and partial hypoxia (5% O2) vis-à-vis normoxia (20% O2, normal oxygen conditions) on the metabolism of glucose in colo-205 colorectal cancer cells in vitro.

POSTER 142
Investigation of the role of hypoxia on Glutamine metabolism in kidney cancer cell via 13C NMR Spectroscopy
Presenting Author: Asiye Asaadzade

Complete Author List:
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Glutamine is the most abundant amino acid in the body and is a major source of carbon and nitrogen . Secondary only to glucose, glutamine is one of the favorite fuels of cancer to sustain. In this study, we have investigated the metabolic fate of glutamine in kidney cancer cells using carbon-13 nuclear magnetic resonance spectroscopy. Due to overexpression of specific glutamine metabolic enzymes, there is an overproduction of glutamine metabolites such as glutamate and alphaketoglutarate which can be detected by 13C. For this project, we have utilized [5-13C] L-glutamine to be able to see the metabolic fate of glutamine, this study will investigate how hypoxia affects the glutamine and glutamate consumption and metabolism under normoxic and hypoxic conditions.

POSTER 143
Extending the Scope of 1H NMR Based Blood Metabolomics for the Analysis of Labile Antioxidants: Reduced and Oxidized Glutathione
Presenting Author: G. A. NAGANA GOWDA

Complete Author List:
G. A. NAGANA GOWDA (University of Washington); Vadim Pascua (University of Washington); Daniel Raftery (University of Washington)

Glutathione is a ubiquitous cellular antioxidant, which is critically required to protect cells from oxidative damage and free radical injury. It is extremely difficult to analyze glutathione in its native form after isolation from biological mixtures since the active form (GSH) spontaneously gets converted to the oxidized form (GSSG). With a focus on addressing this challenge, in this study, we describe an extension to our recent whole blood analysis method [Anal Chem. 2017, 89:4620-4627] that includes the important antioxidants, GSH and GSSG. The presented method broadens the scope of the global metabolite profiling and adds a new dimension to NMR-based blood metabolomics. Further, the method demonstrated here for human blood can be extended to virtually any biological specimen.

POSTER 144
NMR determination of the enantiomeric form of 2-hydroxyglutarate in renal cell carcinoma with an isocitrate dehydrogenase 2 mutation
Presenting Author: Penghui Lin

Complete Author List:
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Biologically important 2-hydroxy carboxylates such as lactate, malate and 2-hydroxyglutarate (2HG) exist in two enantiomeric forms (D and L) with different biological origins and functions and cannot be distinguished under achiral conditions. In order to identify them, we have optimized a derivatization technique using diacetyl-L-tartaric anhydride (DATAN) to produce diastereomers from 2-hydroxy carboxylates enantiomers and applied 1D and 2D NMR methods directly to distinguish the enantiomers without the need for chromatography. We also demonstrated the application of this method on the ex-vivo media and tissue extract of a human renal cell carcinoma and showed the quantitative conversion to the diastereomers, enabling the determination of the enantiomeric ratio of 2-hydroxycarboxylic acids with high precision in various biological samples.

POSTER 145
Radiation Exposure Induces Cross-Species Temporal Metabolic Changes that are Mitigated in Mice by Amifostine
Presenting Author: Alexandra Crook

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Global political unrest has highlighted the importance of understanding γ-radiation exposure to human health and survivability. While the knowledge of nuclear weapons is increasing, our understanding of how to protect and mitigate the effects of radiation injury is still limited. We performed multiplatform metabolomics study to characterize the temporal metabolite changes in animal serum samples following γ-radiation treatment. Three γ-radiation animal model cohorts resulted in 23 consitent dysregulated pathways identified. Amifostine treatment in mice was also evaluated as a radioprotective agent against ARS and showed distinct return to baseline metabolic levels. Our data suggested unique physiological changes can be seen across radiation doses and species through a multiplatform metabolomic approach. Furthermore, a radioprotective signature can be identified through amifostine treatment.

POSTER 146
Targeted Compound Selection in Environmental Samples Using 13C-DREAMTIME in Both High-Field and Low-Field NMR
Presenting Author: Katelyn Downey

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NMR spectroscopy's low sensitivity has long impeded its widespread use in environmental research, as environmental samples are chemically complex and produce poorly resolved spectra. Therefore, this work presents a novel technique called 13C-DREAMTIME, which selectively targets and refocuses desired resonances into singlets in 13C-labelled samples. 13C-DREAMTIME was successfully used at high-field (500 MHz) to select 8 compounds from a 13C-labelled amino acid solution, 6 compounds from 13C-labelled algae, and used at low-field (80 MHz) to analyze a mixture of 13C-L-phenylalanine and 13C-D-glucose. It was also used to monitor metabolic changes in microplastic-exposed Daphnia magna (500 MHz) and track production of ethanol via 13C-D-glucose fermentation (80 MHz). 13C-DREAMTIME's ability to improve sensitivity and resolution holds great promise for future environmental research.

POSTER 147
A Comparison of Methods for Producing Low-Magnetic Susceptibility Micro-coils
Presenting Author: Vincent Moxley-Paquette

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Previously, 5-axis CNC micro-milling was shown to be a viable alternative to traditional micro-coil production methods and was used to create a prototype Slotted-Tube Resonator (STR). Although an excellent Limit of Detection was shown, the lineshape was reduced due to the magnetic susceptibility of the Copper resonator. Here, an Aluminum STR (~291 µm thick) was produced using a MiRA6 5-axis CNC milling machine and electroplated with various thicknesses of Copper (each metal having an opposing magnetic susceptibility) ranging from 4.5 µm to 15 µm. Lineshape improved as the Copper thickness reached 12.5 µm before deteriorating. In this presentation,

multiple methods will be employed to reduce the impact of magnetic susceptibility on lineshape, including the use of both Alloys and electroplating.

POSTER 148
Cutting Without a Knife – a 2D Slice Selective NMR Technique for the Analysis of Inhomogeneous Samples
Presenting Author: Daniel Henryk Lysak

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Despite nuclear magnetic resonance (NMR) being a powerful analytical technique, solution state NMR struggles with heterogenous samples (e.g., studying phase transfer catalysis, or in-vivo organisms). To address this, we introduce a novel 2D, slice selective 1H-13C heteronuclear single quantum coherence (HSQC) sequence that employs pulse field gradients to examine selected “slices” within an NMR tube, and demonstrate its applicability to a three-layered organic chemistry sample, a micro-coil array, and a living worm (Eisenia fetida), before and after feeding with 13C labelled algae (Chlamydomonas reinhardtii). Overall, it was found that this technique gives artefact free slice selection, can aid in the study of multi-layered or heterogenous samples, and shows potential in examining toxicity and bioaccumulation in specific tissue regions, in-vivo.

POSTER 149
Microfluidic NMR for In Situ Culture and Metabolomic Analysis of Hepatocellular Carcinoma Cell Lines
Presenting Author: Evie Rogers

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This work demonstrates the use of microfluidic NMR for in situ culture and quantitative analysis of metabolism in hepatocellular carcinoma cell lines. Building upon previous metabolic data with 4-8 hour time resolution, this work provides quasi-continuous NMR observation for a more detailed view of cellular metabolism and how it reacts to external stimuli. This technique is non-destructive, non-invasive and can measure mM concentrations at µl volumes, within a few minutes and in precisely controlled culture conditions. This is sufficient to observe changes in primary energy metabolism, using only 1,500 - 2,000 cells per device, and with data points every 17 minutes for 24 hours. This has significant potential in the pharmaceutical field for drug development, particularly in toxicity testing.

POSTER 150
Investigation of Brain Tissue Metabolite Local Environment with HR-MAS
Presenting Author: James Collins

Complete Author List:
James Collins (University of Florida)

Spectra obtained using HR-MAS approaches the line-widths of those seen in solution state NMR experiments, and allows for over a dozen small molecule metabolites to be identified. The local environment of the metabolite, such as their presence in cytosol, extracellular space, cell membranes, etc., lead to changes in their molecular dynamics. Preserving some of this structure in the excised tissue, allows for these interactions to be studied using techniques that are sensitive to molecular motions, such as Relaxometry and Diffusion measurements. PROJECT based sequences were used to acquire data regarding molecular motions in excised tissue. It is proposed that such sequences can be modified to provide a single angle of a double diffusion encoded sequence, providing additional micro-structural information.

POSTER 151
Hyperpolarized [2-13C]Pyruvate as a Probe of Hepatic Ketogenic Capacity
Presenting Author: Matthew Merritt

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Ketone production is a metabolic safety valve that can protect tissues from oxidative damage, and is also a major source of energy in fasting conditions. Here we demonstrate that hyperpolarized [2-13C]pyruvate can be used to directly observe ketogenesis in the perfused mouse liver. Livers were perfused in control conditions, and in two conditions known to amplify ketone production. Carbon-13 spectra were acquired at 14.1 T after injection of the pyruvate substrate, and showed production of acetoacetate and beta-hydroxybutyrate. Production of ketones was elevated in both experimental conditions. Gas chromatography- mass spectrometry analysis of the extracted livers and the perfusate revealed strong 13C enrichment of the ketones, confirming the chemical shift assignments in the 13C spectra.

POSTER 155
Design and Construction of A Double-Tuned RF Coil for 1H/31P Solid State MRI at 1.5 T
Presenting Author: Jingting Yao

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A double-tuned single-coil RF probe was designed to acquire solid state 1H and 31P MRI scans of phantoms and animal tissues using a specialized 1.5 T extremity scanner. This unique RF coil has a much larger volume than a typical double-tuned single-coil for spectroscopy. Laboratory tests including circuit simulations, VNA characterization and MR imaging demonstrated the feasibility of this coil in the application of 1H/31P for liquid phantoms. Upon optimizing B1 homogeneity by adjusting the turn spacing, this coil will be used for interleaved 1H/31P acquisition and cross-polarization pulse sequences for solid materials. The MR information on organic matrix (1H) and bone mineral density (31P) may contribute to a more informative diagnostic and effective therapeutic intervention of metabolic bone disease.

POSTER 158
Practical aspects of correcting systematic errors on fiber tracking of selected brain ROIs.
Presenting Author: Anna Stefańska-Bernatowicz

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Anisotropy can facilitate detection of brain tissues pathology, and neurodegenerative disease. It can inform about brain asymetries. Magnetic field gradients supposed to be constant in space for the examination object in all of directions. The lack of homogeneity can have influence to the b-matrix and consequently affects the systematic errors of the DTI images. Calibration of the b-matrix based on diffusion gradient correction in all directions with phantoms, can be use to eliminate those errors. BSD-DTI can help to define precisely value of FA and MD and improve the image accuracy of fiber tracts. Aim of the study was to reveal progression of BSD-DTI method to improved visualization of anisotropic structures of human brain on the example of Uncinate Fasciculus.

POSTER 159
Moving MRI: Imaging a Moving Body with a Moving MRI Magnet
Presenting Author: Jingting Yao

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This work introduces Moving Magnetic Resonance Imaging (mMRI), presenting one implementation of this technology. The ultimate goal is to develop a prototype mMRI scanner in which the magnet and the subject together undergo substantial motion (rotations, translations, and tilts) while remaining stationary with respect to each other. Brain imaging during naturalistic motion may reveal how the brain senses and controls bodily motion, balance and homeostasis. Moving the magnet concurrently with the subject minimizes motion artifacts and field-induced physiological effects. The outcome of this work may improve understanding of brain disorders that are revealed only while the head and/or body are in motion, and may shed light on how the brain suffers and recovers from traumatic brain injury.

POSTER 161
Parallel Ultra-low Field MRI with a Multichannel Optically Pumped Magnetometer
Presenting Author: Young Jin Kim

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Magnetic resonance imaging (MRI) at ultra-low field (ULF) (<<1 T) is a promising non-invasive method for anatomical imaging. Previously, multichannel ULF MRI was realized only with multiple high-sensitivity superconducting quantum interference devices (SQUID). A serious drawback of SQUID sensors is the need for cryogenic infrastructure. To address this limitation, we have designed an ambient-temperature multichannel sensor composed of a multichannel optically pumped magnetometer (OPM) coupled to multiple flux transformers (FTs). In this talk, we will report our progress on the development of the multichannel FT-OPM ULF MRI sensor. In addition, we will present our recent ULF MRI experiment based on a specially designed pick-up coil that achieves 1 mm resolution images of a phantom in about 5 minute scan time.

POSTER 164
K Space Trajectories for Imaging in a Single Sided Low Field MRI
Presenting Author: Muller De Matos Gomes

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There is a need to expand the accessibility of MRI. Doing so would allow physicians to detect illnesses earlier, which would allow for earlier treatments, which could result in better clinical outcomes. Low field MRI scanners, small enough to be placed in a doctor's office, would allow for more routine MRI scans, making it possible to achieve better clinical outcomes from earlier detection. Reducing the size of an MRI scanner typically comes at the cost of making its main magnetic field weaker and more inhomogeneous, making it necessary to radically alter the design of imaging pulse sequences. K space trajectories for imaging in an inhomogeneous field with chirped pulses are developed for single sided low field systems.

POSTER 165
Resolving Chemical and Spatial Heterogeneities at Complex Electrochemical Interfaces in Li-Ion Batteries
Presenting Author: Lauren Marbella

Complete Author List:
Lauren Marbella (Columbia University)

Here, we use solid-state nuclear magnetic resonance (SSNMR) to provide chemical and spatial information, on the nanometer length scale, on the cathode electrolyte interphase (CEI) deposited on LiNi0.8Mn0.1Co0.1O2 (NMC811) composite cathode films in lithium ion batteries. Paramagnetic interactions (assessed with electron paramagnetic resonance (EPR) and relaxation measurements) in 13C SSNMR provide information on the radial arrangement of the CEI from the NMC811 particles outward. Using this approach, in conjunction with X-ray methods, we find that LiF, Li2CO3, and carboxy-containing structures are directly appended to NMC811 active particles, whereas soluble species detected during in situ 1H and 19F solution NMR experiments (e.g., alkyl carbonates, HF, and vinyl compounds) are randomly deposited on the composite surface.

POSTER 166
Characterization of Polymer Molecular Weight Distribution by NMR Diffusometry: Experimental Criteria and Findings
Presenting Author: Jianbo Hou

Complete Author List:
Jianbo Hou (Dow Inc.); Eric Pearce (Dow Chemical)

NMR diffusion is a powerful tool to characterize molecular weight (M) and molecular weight distribution (MWD) of polymers. Despite many published results on this topic, data disparity exists. We present a fundamental study to show how the measurement condition impacts the determined M/MWD. We use the critical dilute concentration C*dilute to explicitly delineate the boundary of the sufficiently dilute condition, below which chain interactions are negligible. We present solid evidence to validate the postulated theory that links C*dilute to M, polydispersity and chain conformation. These findings provide useful guidance for M/MWD characterization by NMR

diffusion. It further sheds light on the interplay between chain conformations and diffusion for globular structure and offers a new approach toward characterizing architecture and M.

POSTER 168
Studying CO2 Speciation and Dynamics in Amine-modified Mesoporous Silica by CSA and Relaxation 13C NMR
Presenting Author: Ildefonso Marín-Montesinos

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Global warming has triggered the development of CO2 capture technologies from large point emission sources as one of the most efficient ways to mitigate CO2 emissions. Amine-modified mesoporous silicas (AMPS) have attracted major attention as the next generation of CO2 adsorbents as their surfaces can be easily tailored for improved CO2 adsorption. Therefore, CO2 adsorption mechanisms and chemical speciation in AMPS has been intensively researched. Our group has successfully applied a combined solid-state NMR and modeling approach to address CO2 chemisorption mechanisms in AMPS.
In this presentation, for the first time, we demonstrate how relaxation and CSA 13C solid-state NMR measurements can be used to fully quantify and characterize the dynamics of confined chemisorbed and physisorbed CO2 species in AMPS.

POSTER 169
A 55Mn ssNMR Investigation of Manganese Dioxide Polymorphs
Presenting Author: Anne Mirich

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To date, solid state NMR (ssNMR) on materials containing paramagnetic species have been performed on reporter ions like 7Li, 19F, 27Al, 29Si. However, many metal oxides do not contain these ions or nuclei in their structures. To establish typical ssNMR spectra of a representative metal oxide framework, 55Mn NMR was performed on various MnO2 polymorphs. Combined with powder XRD, HRTEM, EPR, and Raman scattering spectra, different manganese oxide phases were fully characterized. ssNMR revealed the bulk of the transition metal oxide, giving rise to spectral signals not observed in XRD. HRTEM was used to confirm the surface and tunnel structure of select MnO2 polymorphs in comparison to ssNMR spectra, specifically electrochemical manganese dioxide (EMD) and nsutite.

POSTER 170
NMR as a node of convergence in molecular water science
Presenting Author: Eric Breynaert

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Unique, but fragmentary information on self-organization of water in its liquid form, at surfaces, and around biomolecules, is being harvested by physicochemical observations and a score of state-of-the-art techniques, Nuclear Magnetic Resonance (NMR), Electrochemical Impedance Spectroscopy (EIS), Dielectric Relaxation Spectroscopy (DRS), infrared, Raman and TeraHertz spectroscopy as well as X-ray and neutron scattering. Probing chemistry, chemical interactions and physical state, NMR is ideally positioned to serve as a node of convergence connecting observations from different sources into a unified model. This contribution will show, using examples of real chemical processes how NMR spectroscopy excels in this role.

POSTER 172
Influence of cation structure on molecular properties of ammonium ionic liquids
Presenting Author: Adam Klimaszyk

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This work aimed to characterize microscopic interactions by terms of self-diffusion coefficient and evaluate the microscopic and macroscopic structure of three ammonium ILs' families, choosing cations of varied structure (aromatic, cyclic, and aliphatic), varied alkyl chain length attached to the quaternary nitrogen, and bis(trifluoromethanesufonyl)imide anion, which results in three homologous series of ionic liquids with the cation of different geometry, with different physicochemical properties.
After structure confirmation through 1H NMR and FTIR, phase transition temperatures were determined using differential scanning calorimetry (DSC). Self-diffusion coefficients were determined for a series of the alkyltriethylammonium ILs measurements at 14,4 T Agilent NMR spectrometer techniques.

POSTER 173
NMR as a Discovery Tool: Searching for the Unexpected in Industrial Effluents Discharged into the Environment
Presenting Author: Kiera Ronda

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Ontario's continued industrial growth has contributed to increasing concerns surrounding the environmental implications of industrial effluents. These concerns have emphasized the need for revised regulations and detection methods. Traditional methods rely on targeted mass spectrometry, which is limited to the detection of known pollutants. NMR spectroscopy offers an ideal solution, as it allows for the identification of previously unknown contaminants. This study will analyze wastewater samples from nine of Ontario's industrial sectors to compile a comprehensive database of chemical pollutants. This will include qualitative and quantitative data to provide a detailed analysis of potentially harmful environmental pollutants. The resulting database can then serve as a foundation for updated regulations surrounding the release of potentially harmful substances.

POSTER 174
Molecular-Level Insight of Phase stability in Phase-Change Nano-Emulsions Stabilized by Stearic acid for Thermal Energy Storage
Presenting Author: Jungeun Park

Complete Author List:
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Phase Change Materials (PCMs) are latent heat storage materials that can store or release thermal energy during phase transitions. Organic PCMs can be emulsified in water in the presence of surfactants to improve thermal conductivity and transport properties. However, PCMs nano-emulsions can becomes unstable during thermal cycling in heat transfer systems. To better understand the molecular behavior of PCMs nano-emulsions, solution-state NMR were applied to a PCM nano-emulsion containing octadecane, water, and 13C-enriched stearic acid as a surfactant. Solution-state 1H and 13C NMR methods observed freezing and melting of octadecane nano-emulsion, including supercooling effects, as well as multiple chemical environments of the surfactant head group. 1H Rheo-NMR experiment were applied to measure the flow behavior of PCM nano-emulsion.

POSTER 175
Calcium-43 and Oxygen-17 solid state NMR studies of biomaterials: From isotopic labeling to ultra-high field NMR analyses and DNP
Presenting Author: Danielle Laurencin

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Oxygen and calcium are highly abundant elements in living organisms.
Here, some of our recent work aiming at using 17O and 43Ca NMR to investigate the structure of synthetic biomaterials will be presented. For 43Ca: the optimized synthesis of 43Ca-enriched phases relevant to bone mineral will be described, as well as characterizations at ultra-high magnetic fields (1.5 GHz) and using dynamic nuclear polarization. For 17O: innovative labelling schemes for the preparation of 17O-enriched hydrates using mechanochemistry will be presented, and it will be shown how these can be used for reaching insight into the dynamics of water molecules.
For both nuclei, the importance of using combined experimental-computational approaches will be highlighted.

POSTER 176
Synthesis, Characterization, and NMR Crystallography of Multi-Component Crystals of Urea
Presenting Author: Cameron Vojvodin

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NMR crystallography based on NMR spectra of quadrupolar nuclei (i.e., QNMRX) can be used to solve, refine, and validate crystal structures. In this

POSTER, we investigate multi-component crystals (MCCs) of the form NR4Cl:xUrea·yH2O made by ball milling, their characterization by 35Cl SSNMR and PXRD, and the use of ab initio molecular dynamics calculations to study the impact of molecular motions on 35Cl EFG tensors for site assignments and QNMRX. Additionally, we apply our QNMRX method to investigate MCl:Urea:xH2O MCCs made by ball milling, which are characterized by 35Cl, 23Na, 7Li, and 133Cs SSNMR, synchrotron XRD, and thermogravimetric analysis. Plane-wave DFT calculations and Rietveld refinement of synchrotron XRD data aid in determining the crystal structures of these model systems.

POSTER 177
Mechanochemical Syntheses of Pharmaceutical Cocrystals and their Structural Characterization Using 35Cl Solid-State NMR and Dispersion-Corrected DFT Calculations
Presenting Author: Zachary Dowdell

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The solid forms of active pharmaceutical ingredients, such as polymorphs, cocrystals, and hydrates, have distinct physicochemical properties. HCl salts of APIs are the most common solid forms; however, their physicochemical properties can often be improved via synthesis of pharmaceutical cocrystals (PCCs) with appropriate coformer molecules. Herein, we describe the use of 35Cl solid-state NMR spectroscopy for probing the structures of promethazine HCl and chlorpromazine HCl PCCs. Each PCC has Cl– environment(s) with unique hydrogen bonding networks, and therefore, distinct 35Cl electric field gradient (EFG) tensor parameters, which manifest as different central transition powder patterns. Plane-wave DFT methods are used to refine the structures of PCCs, utilizing comparisons of experimentally-measured and computationally-derived 35Cl EFG tensors as structural evaluation metrics.

POSTER 178
Wine Making 101: An Undergraduate Experiment Utilizing Low-Field NMR for Quantification and Process Monitoring
Presenting Author: Amy Jenne

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Undergraduate students, while taught NMR theory, often are unable to gain hands on experience. In this research, a novel undergraduate laboratory experiment is created that leverages the affordability and useability of benchtop NMR, currently underutilized as an educational tool. Students are asked to follow the process of alcohol fermentation by using ERETIC to quantitatively compare the growth of Ethanol and the decay of D-Glucose. With quantitative measurements, students will be able to measure the kinetics of this

process relating lab experiences to their in-class learning. This experiment offers new tools for undergraduates with practicality beyond their degrees, coupled with the affordability of a benchtop NMR as an instrument for education.

POSTER 179
Automate High Throughput NMR to Support Parallel Medicinal Chemistry in Drug Discovery
Presenting Author: Ronghui Zhou

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Highly efficient access to a large variety of analogs is critical to the success and timeline of hit identification and lead optimization in modern drug discovery. Parallel synthesis is extensively exploited in this regard to rapidly prepare hundreds of analogs in library format to reduce the Design-Make-Test cycle time. Within the Chemical Capabilities, Analytical and Purification organization at the Janssen Global Discovery Chemistry, our High Throughput Purification and Analytical Chemistry teams are working closely with the Parallel Medicinal Chemistry team to rapidly purify these libraries via mass-directed reversed-phase purification, which is then followed by structural verification by high-throughput NMR. In this work, we will present our semi-automated high-throughput NMR workflow, from sample preparation, data acquisition to structure verification and reporting.

POSTER 180
Use of Diethanolamine as Viscous Solvent for Mixture Analysis by Heteronuclear ViscY NMR Experiments
Presenting Author: LEROY

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The use of diethanolamine/DMSO-d6 as viscous binary solvent is reported for the individualization of low-polarity mixture components by heteronuclear ViscY NMR experiments under spin diffusion conditions. Solvent viscosity induces the slowing down of molecular tumbling, hence promoting magnetization transfer by dipolar longitudinal cross-relaxation. As a result, all 1H nuclei resonances within the same molecule may correlate in a 2D NOESY spectrum, giving access to mixture analysis. We state the individualization of four low-polarity chemical compounds dissolved in diethanolamine/DMSO-d6 solvent blend using homonuclear selective 1D, 2D 1H NOESY and 19F HOESY experiments and heteronuclear 2D 1H-19F, 1H-31P HSQC-NOESY and 3D 1H-19F-1H, 1H-31P-1H HSQC-NOESY experiments by taking profit from spin diffusion.

POSTER 181
Selective excitation of overlapping multiplets
Presenting Author: Peter Kiraly

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GEMSTONE is a recently described methodology for the selective excitation of overlapping multiplets in 1H NMR spectra. Unlike earlier chemical shift selective filter (CSSF) methods, GEMSTONE can achieve this high level of selectivity within a single scan, resulting in a considerable reduction in experiment time, thus broadening the scope and applicability of selective 1D experiments. Here we illustrate GEMSTONE using the NOESY and TOCSY experiments as examples and report some of the important practical and theoretical aspects that underpin the method.

POSTER 182
Characterization and Investigation of a MOF-Based (IrIMes@NU-1000) Catalyst for Hyperpolarization via Heterogeneous SABRE-SHEATH
Presenting Author: Shahabuddin Alam

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We investigate a novel heterogeneous SABRE (HET-SABRE) catalyst based on metal-organic frameworks (MOFs). We use a tunable MOF with adjustable pore size and high surface area, NU-1000, to immobilize the IrIMes moieties. A variety of techniques were used to characterize the MOF HET-SABRE catalyst, including SEM, EDX, and BET. Up to ~7100-fold 15N enhancement was achieved for 15N-acetonitrile via SABRE-SHEATH. This 15N enhancement (P15N~2.4%) would represent a ~102-fold improvement over previous results; however, leaching of catalytic moieties may be more of an issue. We are currently working to improve the catalyst and test it on a wider range of substates.

POSTER 183
Proton NMR spectroscopy based chemosensory differentiation of anti-obesity phytocompounds and medicinal plants
Presenting Author: Ankita Singh

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Obesity is now a major global epidemic leading to diseases such as hyperlipidemia and type 2 diabetes. Recently, newer approaches using polyphenolic phytocompounds and medicinal plants present an exciting opportunity for the development of newer therapeutics. In this study, twenty-two phytocompounds and thirty-eight medicinal plants containing these phytocompounds from sweet and pungent chemosensory groups were studied using proton NMR spectroscopy. Multivariate analysis of NMR data demonstrated the potential of proton NMR in differentiating medicinal plants and phytocompounds from both chemosensory taste groups. Furthermore, anti-lipase assays of medicinal plants have confirmed anti-lipase activity for the pungent category and no anti-lipase activity was seen for sweet category plants.

POSTER 184
Molecular Structures of Reaction Products in $LiPF_6$ Carbonate Electrolyte with a Phosphorous Pentoxide Scavenger for Rechargeable Lithium Metal Batteries
Presenting Author: Leo W. Gordon

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Lithium metal batteries currently suffer from poor cyclability in existing $LiPF_6$ carbonate-based liquid electrolytes due to autocatalytic HF formation in the presence of water or labile protons, which degrades the solid electrolyte interphase (SEI) and dissolves electrode transition metals. Recently, we reported P_2O_5 as a low-cost additive that scavenges both water and HF to mitigate these deleterious side reactions, which also promotes formation of a stable phosphorous-containing SEI layer that promotes remarkably stable electrochemical cycling. However, the detailed reaction mechanisms and products are not known. Here, we elucidate the reaction mechanisms and products formed upon P_2O_5 modification for the first time via liquid-state NMR spectroscopy, yielding insights into how P_2O_5 significantly enhances electrochemical performance.

POSTER 185
Structural Determination of an Unusual Carbanion Process Impurity using NMR, HRMS and X-Ray Crystallography
Presenting Author: Qingmei Ye

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An unknown process impurity (0.7%) with a molecular formula of C24H35FN2O6S3 was observed during the synthesis of a regulatory starting material. It was isolated for NMR structure elucidation, a critical piece of knowledge to support the process validation campaign. The 1D and 2D NMR data were sufficient to assign most of the protons and carbons except one carbon with a chemical shift at 86 ppm that was observed in 1H-13C HMBC, but not in 1D 13C spectrum. This carbon was assigned as an (alkoxysulfonyl)(methylsulfonyl)((triethylammonio)methyl)sulfinyl)methanide anion by single crystal X-ray crystallography. This presentation describes the unusual carbanion NMR data as well as HRMS and single crystal X-ray crystallography. The rationale for the formation of the impurity is also presented.

POSTER 186
Revealing the Conformational Variability of Modified Macrocyclic Peptides by exact NOEs
Presenting Author: Simon Ruedisser

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The pharmacological properties of cyclic peptides and peptidomimetics are strongly influenced by conformational states present in polar and apolar environments. The knowledge of the conformational landscape is therefore key for rational drug design. Exact characterization of the conformational space using NMR has been challenging due to very weak NOE effects for small molecules and the semi quantitative nature of the ROE. Here we describe a novel method of using exact NOEs (eNOEs) for ensemble conformation determination for cyclic peptides by using a combination of technological improvements: high viscosity solvents, novel NMR methodology and improved methods for structure calculations. The method has been applied to the immunosuppressant cyclosporin A and the fungal natural product Omphalotin A.

POSTER 187
When does Computer-Assisted Structure Elucidation (CASE) spare you from recording (IN)ADEQUATE?
Presenting Author: Dimitris Argyropoulos

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INADEQUATE and 1,1-ADEQUATE experiments are the only means by which a scientist can directly observe the carbon skeleton of a molecule. They are considered the “holy grail” of NMR experiments for structure elucidation. However, they suffer from inherent low sensitivity. Computer-assisted structure elucidation (CASE) has now become powerful enough to beg the question: Can CASE eliminate the need for time-consuming, low-sensitivity NMR experiments for structure elucidation? We present an analysis of published structure elucidations that claimed to require (IN)ADEQUATE data and use some of these examples to illustrate when and how structure elucidation can be accelerated with the use of CASE.

POSTER 188
Isotopic Labeling and MA’AT Analysis of O-Glycosidic Linkage Conformation in Complex-Type N-Glycan Oligosaccharides
Presenting Author: Mi-Kyung Yoon

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aMan(1→3)-[aMan(1→6)]-bManOCH3 (1) was converted chemically or enzymatically to b[13C]GlcNAc-(1→2)-aMan-(1→3)-[aMan-(1→6)]-bManOCH3 (2), b[13C]GlcNAc-(1→2)-aMan-(1→6)-[aMan-(1→3)]-bManOCH3 (3), bGlcNAc-(1→2)-aMan-(1→3)-[bGlcNAc-(1→2)-aMan(1→6)]-bManOCH3 (4), and bGal-(1→4)-bGlcNAc-(1→2)-aMan-(1→3)-[bGal-(1→4)-bGlcNAc-(1→2)-aMan(1→6)]-bManOCH3 (5). NMR spin-couplings were measured in 2–5, and trans-glycoside values used to determine phi (H1’–C1’–O1’–C2) and psi (C1’–O1’–C2–H2) in the a-(1→2) linkages of b[13C]GlcNAc-(1→2)-aManOCH3 (6), 2 and 3 using MA’AT analysis. In 6, mean values of 33.5o and 26.9o, CSDs of 34.4o and 24.4o, and RMSDs of 0.06 Hz and 0.18 Hz were found for phi and psi, respectively. Mean phi values were 38.7o (2) and 33.7o (3), and mean psi values were 23.1o (2) and 24.5o (3). CSDs suggest more restricted librational motion about phi in 2 than in 3 and 6. [NSF CHE 1707660 and 2002625]

POSTER 189
A Simple IPAP NMR Experiment to Quantify 13C Enrichment at Low and High Field
Presenting Author: Katrina Steiner

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Green algae transform atmospheric carbon dioxide into organic molecules which are ultimately assimilated and transformed by higher trophic level organisms. The novel in-phase anti-phase (IPAP) experiment presented has potential applications in 13C food web tracing studies. This experiment produces separate 1H NMR spectra based on 1H-12C and 1H-13C connectivity. By comparing these spectra to a reference containing both 12C and 13C signals, absolute quantification can be achieved. A lab-constructed photobioreactor is

additionally introduced which is used to cultivate 13C enriched algae. The presentation will further introduce the NMR pulse sequence, demonstrate its ability to evaluate offline and real-time enrichment, quantify standard mixtures and used to follow biological processes at both high (500 MHz) and low field (80 MHz).

POSTER 190
In-situ Chemical Reaction Monitoring by NMR in Highly viscous solvent under Spin Diffusion Condition
Presenting Author: Francois PEDINIELLI

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The rapid analysis of complex mixtures whose composition evolves with time represents a considerable challenge for the industry and academic stakeholders. Such an investigation is motivated by the understanding of the mechanisms involved in the formation of the final products, which often goes through one or more intermediates. The ViscY (Viscosity-enhanced spectroscopY) approach is a new way to solve this issue. Indeed, the use of viscous solvents offers a new possibility to analyze complex mixtures of time-varying composition. Successfully slowing down the molecular rotation rate of the molecules in a mixture allows to control the reaction dynamics and to group the NMR peaks according to the compounds from which they originate by spin diffusion studies, as revealed by NOESY spectra.

POSTER 191
NMR Spectroscopic Investigation of Acridone-based Interpenetrated Coordination Cages
Presenting Author: Andre Platzek

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From the vast variety of structures derived from supramolecular assembly, coordination cages formed by banana-shaped ligands and Pd(II) cations are of special interest in our group. Some of these Pd2L4 cages based are known to form interpenetrated double cages with three distinct cavities that can incorporate anionic guest molecules as well as neutral guest molecules. Aside from the cavities, the outside and the shape of the cage was of special interest. The effect of different alkyl sidechain lengths R (H3C−(CH2)n with n = 1,3,…,11) in solution was investigated by diffusion ordered NMR spectroscopy (DOSY). To understand the well-known effect of convection during the measurement we compared different pulse sequences on their abilities to compensate the convection.

POSTER 192
HR-MAS Analysis of Skin Exposed to Chemical Warfare Agents
Presenting Author: David J. McGarvey

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In the event of a chemical warfare agent (CWA) release, there is a likelihood of CWA deposition on the exposed skin of an unprotected population. Therefore, it is vital to understand the interaction of CWAs and skin to design and evaluate successful decontamination techniques and technologies.
This research uses HR-MAS NMR to analyze skin samples before and after exposure to chemical warfare agents. The degradation of the agents was followed using this non-destructive technique, and breakdown products and the rates of degradation were measured. Nerve agents and blister agents were investigated. The effectiveness of decontamination methods at different time points was investigated.

POSTER 193
NP-MRD, The Natural Products Magnetic Resonance Database
Presenting Author: John R. Cort

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The Natural Products Magnetic Resonance Database (NP-MRD, np-mrd.org) has been established with a goal to become a comprehensive, searchable, connected, and open database and repository for all natural products NMR data. The mission of NP-MRD is to benefit research through engagement and partnership with the worldwide natural products community. With derived (e.g. chemical shift assignments), raw (FID), and simulated NMR data, as wells as tools and links to other databases, NP-MRD can facilitate dereplication, support correction of erroneous or missing chemical shift assignments, and enable structure validation or structure revision. Furthermore, NP-MRD can create opportunities for developing new artificial intelligence-based approaches for structure determination and chemical shift or spectral prediction, among other presently unforeseen applications of such a database resource.

POSTER 194
Intermolecular Dipolar Cross-Relaxation of Nano-Confined Fluids
Presenting Author: Stacey M. Althaus

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In fluids confined in nanopores, we have found the intermolecular cross-relaxation rates between confined fluid molecules or between fluid and pore matrix are two orders of magnitude larger than those in bulk states. The sign of the cross-relaxation rate is also negative, in contrast to the positive intermolecular relaxation rates in bulk state. The classical NMR relaxation theory fails to capture these observations in a nano-confined fluid environment. A formal theory was proposed for intermolecular cross-relaxation of fluids in nanopores by accounting for the nanoconfinement to the translational diffusion of the fluid molecules. The calculated results from the theory are consistent with measured intermolecular cross-relaxation data between different liquid molecules in a nanopore or between liquid and nanopore matrix.

POSTER 195
Pure Isotropic Proton Solid State NMR
Presenting Author: Bruno Simões de Almeida

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Homonuclear dipolar couplings contribute the most to the broadening of 1H spectral lines in solid-state NMR. Magic-angle spinning (MAS) improves spectral resolution and, at the highest rates available (100-150 kHz), linewidths are reduced to hundreds of hertz. However, due to the imperfect nature of coherent averaging, the interactions are not averaged out
Completely and the residual linewidths are still orders of magnitude broader than those encountered in NMR of liquids.
Here, we propose, instead of optimizing an averaging scheme, to parametrically map the residual terms deriving from the imperfect averaging of MAS and remove them in a k-space representation. Tested on eight different organic solids, we obtained isotropic spectra 20 times narrower than the 100 kHz MAS spectra.

POSTER 196
A Versatile MRI Post-Processing Package with Graphical User Interface in MATLAB
Presenting Author: Victor Kassey

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Magnetic Resonance Imaging is one of the potential medical diagnostic modalities used in healthcare. Image post-processing is important in quantitative data analysis. Magnetic field (B0), radiofrequency (B1), susceptibility, motional, T1/T2 relaxation times cause image distortions in MRI. These distortions are resolved by post-processing algorithms with improved image quality, signal-to-noise ratio, and contrast uniformity. There is a need for a versatile post-processing package to resolve disparities since the commercial packages have limited capabilities. The present package is developed to perform image post-processing and data analysis with accurate auto-and-manual segmentation/registration with B1/B0 correction capabilities. The B1/B0 corrections can be implemented either with homogenous phantom data or bias field correction. It is a fuzzy-C-means (FCM) based package developed in MATLAB with GUI capabilities.

POSTER 198
Zero- to Ultra-low Field NMR: the Future of Cheap and Portable Magnetic Resonance
Presenting Author: Adam Ortmeier

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Zero- to Ultra-low Field (ZULF) NMR is a relatively new and exciting technology that–when paired with hyperpolarization techniques–can help in decreasing cost, increasing sensitivity, and introduce portability to magnetic resonance technology. With this technology comes a great deal of theory that has yet to be explored. Here we will show a ZULF NMR setup and explain how it works. We will also talk about how the detection axis at ZULF results in different spectra of the same molecule. In order to understand the theoretical components of why this might happen, we must first ask a couple of questions. How does the initial state evolve under the Hamiltonian? What are the coherences that evolve? Our friends Wigner and Eckart can help.

POSTER 199
Neural Network for Lipid Profile Determination from Time-domain NMR Data
Presenting Author: Hayden Johnson

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A neural network approach is proposed for determining the quantitative NMR lipid profile from FID data to reduce operator-bias and increase speed in NMR lipidomics data processing compared to conventional frequency-domain and time-domain Bayesian approaches. A synthetic data generation workflow is presented to produce 300,000 FIDs with varied combinations and concentrations of several lipid species from 14 original scans of individual lipid species. The trained network was applied to a dietary metabolomics dataset and produced similar statistical results compared to Bayesian analysis, revealing increased hepatic lipid levels in high fat fed compared to chow fed animals and higher lipid levels in females than males. The network approach increased automation and decreased quantification time from 2-3 hours to under 2 seconds.

POSTER 200
Workflow Tools to Evaluate and Optimize Advanced Nonuniform Sampling Methods
Presenting Author: D. Levi Craft

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Nonuniform sampling (NUS) has allowed spectroscopists to tailor their experiments to reduce data collection time and improve spectral quality. We explore an emerging approach to NUS called partial-component sampling (PCS). The Nonuniform Sampling and Reconstruction Contest (NUScon) has released a workflow for evaluating spectral reconstructions. We extend this workflow to accommodate PCS and we add a new evaluation metric using \textit{in situ} receiver operating characteristic (IROC). Together, these new tools provide the infrastructure to optimize PCS experiments for complex biomolecules.

POSTER 201
Direct Optimization of CPMG Refocusing Cycles for Robustness in Time-Dependent B0 Fields
Presenting Author: Soumyajit Mandal

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The time evolution of the Carr-Purcell-Meiboom-Gill (CPMG) pulse sequence in time-varying B0 fields is controlled by an effective Hamiltonian that is a function of the offset field and sequence parameters. The spins adiabatically follow this effective Hamiltonian except at certain distinct offsets where nonadiabatic transitions can occur. The question of whether refocusing pulses can be optimized to reduce sensitivity of the CPMG sequence to field fluctuations, is still open; earlier work was limited to simple rectangular refocusing pulses. Here we explore this possibility by using numerical optimization methods. We find that constant-amplitude refocusing pulses optimized for both sufficient SNR and robustness to B0 fluctuations have similar symmetric phase profiles that are largely independent of length, modulation rate, and field homogeneity.

POSTER 202
Hyperfine Decoupling of Magnetic Resonance Spectra Using Wavelet Transform
Presenting Author: Madhur Srivastava

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The objective of spectral analysis is to resolve and extract relevant features from experimental data in an optimal fashion. We exploit the multiresolution property of wavelet transforms that allow the separation of distinct features of a spectrum based on simultaneous analysis of spectrum and its varying frequency. We retain the wavelet components that stored the hyperfine and/or super-hyperfine features, while eliminating the wavelet components representing the remaining spectrum. Using cw-ESR, we tested the method on simulated cases of metal-ligand adducts and an experimental case of a copper(II) complex with distorted octahedral geometry. The method was able to extract g and hyperfine coupling constant values, and revealed features that were buried in the overlapped spectra.

POSTER 203
Making the Most of Phase-Cycled Data: A New Schema for Organizing, Processing, and Presenting Coherence Transfer Pathways
Presenting Author: Alec Beaton

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A comprehensive, non-standard approach to processing and presenting data from the various coherence pathways accessed by a magnetic resonance experiment is presented. This formalized approach--i.e. "schema"--significantly improves the speed with which spectroscopists can develop new types of experiments. When confronted with adverse experimental circumstances, it guides the identification of data and the development of routines that improve the quality of that data. As the authors' lab has developed an Overhauser Dynamic Nuclear Polarization (ODNP) system, these methods serve here to improve the quality of ODNP data, with particular focus on mitigating the effects of inhomogeneity and time instability of the fields offered by conventional room temperature electromagnets. Relevance to 2D spectroscopy and standard density matrix simulations will also be discussed.

POSTER 204
Determining fast chemical exchange rate constants in nanoemulsions using nuclear magnetic resonance
Presenting Author: Zhaoyuan Gong

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In this work, the second-order kinetics of molecules exchanging between chemically distinct microenvironments, such as those found in nanoemulsions, is studied using NMR. A unique aspect of NMR exchange studies in nanoemulsions is that the difference in molecular resonance frequencies between the two phases, which determines whether the exchange is fast, intermediate, or slow on the NMR timescale, can depend upon the emulsion droplet composition, which is also determined by the kinetic exchange constants themselves. Within the fast-exchange regime, changes in resonance frequencies and line widths with dilution were used to extract the exchange rate constants from the NMR spectra. As a demonstration, the kinetic exchange parameters of isoflurane release from an emulsion made of isoflurane and perflurotributylamine were determined.

POSTER 205
The BMRB archive of Protein, Nucleic Acid and Metabolite NMR Data
Presenting Author: Kumaran Baskaran

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The Biological Magnetic Resonance Data Bank (BMRB: https://bmrb.io) serves the biomolecular NMR community by supporting a curated archive of primary and derived data and metadata linked to scientific investigations under the “FAIR Principles” (Findable, Accessible, Interoperable, and Reusable). BMRB’s goal is to empower scientists in their analysis of the structure, dynamics, and chemistry of biological systems and to support further developments in the field of biomolecular NMR spectroscopy. BMRB uses the NMR-STAR data model, which is defined by a data dictionary. BMRB launched BMRbig (https://bmrbig.bmrb.io) in 2020 to accommodate the acquisition of diverse data (not just NMR data) beyond the types currently curated and annotated by BMRB. BMRB is supported by the US National Institutes of Health.

POSTER 206
Searching Libraries of Known Structures for Dereplication: Benefits and Requirements
Presenting Author: Nadia Laschuk

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Several methods exist to help determine if a newly isolated compound is truly novel or already known. The use of 13C NMR data to search databases of predicted spectra have been previously shown to be beneficial for this. To explore further, we searched a library of predicted 13C spectra using experimental data of 56 compounds. We explored when molecular weight information and consideration of missing or extra peaks in the experimental spectrum were required to identify the correct structure in all cases. Detailed results alongside an optimized workflow that allows one to unambiguously find the correct structures in the database within a few seconds will be presented.

POSTER 207
An Analytical Model for Uncertainty in Longitudinal Relaxation Measurements of Hyperpolarized Media
Presenting Author: Qing Wang

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Highly nonequilibrium nuclear spin polarization (hyperpolarization, HP) can be generated via a range of physical and chemical processes, enabling otherwise impossible spectroscopic and imaging studies. However, HP magnetization decays irreversibly due to RF pulsing with longitudinal relaxation. The underlying longitudinal relaxation rate, r_1, can be of fundamental interest for multiple applications and determines the lifetime of the nonequilibrium in virtually all HP experiments. However, little consideration has been given to the accuracy of the HP relaxation measurement itself. Here, we present a first-principles, analytical model to quantitatively predict the impact of various experimental parameters on the uncertainty of HP r_1 measurements and discuss how this information can be used to improve the accuracy of HP NMR measurements.

POSTER 208
Molecular-Level Understanding of Electrochemical Discharge and Degradation Mechanisms in Li-CFx Battery Electrodes Revealed by Solid-State NMR Spectroscopy
Presenting Author: Loleth E. Robinson

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The NASA Europa Lander mission concept aims to search for signs of life on this moon of Jupiter, which contains water underneath its icy surface. NASA is vetting energy dense Li-CFx battery systems to meet the unique specifications required for the proposed mission. However, much remains to be understood about the electrochemical discharge mechanism at a molecular level, or how radiation affects the local compositions and structures of the electrode materials at different states-of-charge. Through quantitative single-pulse 19F and 7Li solid-state NMR measurements, as well as dipolar-mediated 19F{19F} and 19F{7Li} NMR experiments, we elucidate the molecular-level environments present and quantify the resultant electrode compositions as a function of both state-of-charge and irradiation.

POSTER 209
Unpicking the Neural Networks of DEERNet
Presenting Author: Jake Keeley

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Site-directed spin labelling in EPR yields useful information on distance distributions in biomolecules. However, extracting those distributions is a mathematically ill-posed problem. Tikhonov regularisation – a popular solution – can be fiddly and susceptible to user bias in the selection of regularisation parameters. A recently established alternative is deep neural networks.
This communication reports the technical side of machine learning in DEER spectroscopy: data preprocessing, background signal retrofitting, uncertainty analysis using network ensembles and Jacobians, internal consistency enforcement, detection of corrupted data, dealing with sparsely sampled data, etc. DEERNet is also a rare case of a neural network whose internal functioning is generally understood; we present a behind-the-scenes look at how architectural choices were made to improve the network performance.

POSTER 210
Visual Assist: A Solid State NMR Type and Sequential Assignment Assistance Program
Presenting Author: Tyrone Thames

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A major bottleneck to the usefulness of solid-state NMR experimentation is the data analysis step, wherein chemical shift assignments must be made for as many of the individual atoms in a sample as possible. While for solution NMR experimental data there are automated assignment algorithms that quickly make accurate type and sequential residue assignments, for solid state NMR this task is known for being time-consuming, requiring manual human intervention and analysis, due to the broad linewidth and lower signal-to-noise ratio in solid-state NMR spectra compared to solution NMR.
Presented here is a program for improving residue type assignment by speeding up the process of isolation of resonances belonging to the same residue across NCACX and NCOCX correlated spectra.

POSTER 211
Network for Advanced NMR Data Transport
Presenting Author: Chris Bontempi

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The Network for Advanced NMR (NAN) is an NSF-supported project aimed at simplifying and democratizing the use of high-field NMR spectrometers. The NAN Data Transport System (NDTS) will automatically gather experimental data and link user, project, study, and sample information with it. NAN will include an advanced Data Browser through the NAN web portal for search and retrieval of data across your personal, lab and public data, as well as for managing data sharing.
NAN, with Data Transport, will aid researchers in satisfying NIH grant data sharing plan requirements (NOT-OD-21-013), due to begin in January of 2023.
NAN will also integrate with UCHC's other NMR resources, namely NMRbox for access to hundreds of NMR software and computational clusters, and BMRB.

POSTER 212
Statistically efficient NMR fragment screening
Presenting Author: Hamid R. Eghbalnia

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Ligand affinity screening by NMR is a powerful technique routinely used in drug discovery or functional genomics. Binding events can be identified by examining differences in the 1D 1H NMR spectrum of a compound with and without protein. To improve the efficiency of these screens, compounds are usually evaluated in mixtures of compounds that ideally have non-overlapping peaks. While the non-overlap criterion may detect simple events, it is rarely statistically efficient, in the sense that it is difficult to automate sensitive and specific hit detection. We propose an alternative strategy for creating mixtures that optimize for statistically efficient hit detection. This mixture matrix can include higher counts of compounds in a mixture, and redundantly include compounds among mixture.

POSTER 213
103Rh Solid-State NMR: New Experimental and Theoretical Pathways
Presenting Author: Jasmin Schoenzart

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103Rh is a I = 1/2 nuclide with NMR powder patterns often dominated by the chemical shift anisotropy (CSA) interaction. However, 103Rh has a very low gyromagnetic ratio, γ, which presents challenges for detection of signal and construction of probe circuits free of acoustic ringing. Herein, I will discuss the optimization of 1H-103Rh broadband adiabatic-inversion/cross polarization (BRAIN-CP) methods for the acquisition of 103Rh SSNMR spectra, and show applications of these methods to several Rh coordination compounds at two fields (21.1 T and 35.2 T) using probes adapted for low-γ NMR experimentation. I will also compare experimentally determined rhodium CS tensors with magnetic shielding tensors obtained from DFT calculations on cluster-based models that incorporate spin-orbit relativistic effects and hybrid exchange-correlation functionals.

POSTER 214
NMRbox, a Shared Software and Computation Resource for NMR
Presenting Author: Gerard Weatherby

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NMRbox is a computational platform for NMR data processing and analysis with 240+ software packages for NMR and related fields. NMRbox provides access to software and compute resources via VNC virtual desktops and secure shells (ssh) . Compute resources include 3,000+ CPU cores, 20+ TB RAM, and 200,000+ CUDA cores. NMRbox fosters reproducible research by providing persistent access to pre-configured software, tools for managing the metadata associated with processing workflows, and tools for advanced Bayesian analytics.
Here we present recent enhancements, including continuous releases, system dashboard, user groups, citation management, pre-staged reference data (e.g. AlphaFold, PDB/BMRB), and self-service workshop support.

POSTER 215
NUScon: New Tools for Evaluating Spectral Processing Tasks and New Community Challenges
Presenting Author: Adam Schuyler

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The Nonuniform Sampling Contest (NUScon) is a community-based critical assessment of NUS challenge problems with the objective to determine best practices for processing and analyzing NUS experiments. We address this objective through the following initiatives:

(1) develop quantitative tools for assessing spectral quality; (2) build a modular software package so that each task in a spectral processing workflow can be isolated and evaluated; (3) engage with the NMR community to identify critical challenges; and (4) generate open challenges and gather submissions to identify best practices. We are presenting a new release of the NUScon software package and announcing the NUScon 2022 challenge problems in Sample Schedule Design and Peak Picking.

POSTER 216
Crystal Structure Prediction Based on 35Cl Electric Field Gradient Tensors: New Protocols and Applications
Presenting Author: Carl H. Fleischer III

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Crystal structure prediction (CSP) is an important set of methodologies for generating thousands of candidate crystal structures and selecting the most thermodynamically stable forms. CSP methods are now used with NMR crystallography (NMRX), combining information from solid-state NMR spectroscopy, X-ray diffraction, and quantum chemical calculations for the prediction, refinement, and validation of crystal structures. We have constructed an NMRX/CSP protocol based on 35Cl EFG tensor parameters for simple organic HCl salts. Herein, we discuss these protocols and their applications to problems in crystal engineering, including their: (i) use for prediction of structures of diastereomers that are distinguishable by 35Cl SSNMR spectra; (ii) extension to three-body NMRX/CSP for organic salt hydrates; and (iii) application to a “blind” structural refinement.

POSTER 217
DFT/ZORA Calculations of ^{195}Pt Magnetic Shielding Tensors
Presenting Author: Sean Holmes

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Quantum chemical calculations are invaluable for relating chemical shift tensors obtained from solid-state NMR measurements with molecular-level structure and dynamics. In this

POSTER, we discuss relativistic DFT calculations of ^{195}Pt magnetic shielding tensors. We also consider the calculation of magnetic shielding tensors for light phosphorus atoms bonded to platinum atoms, as well as J-coupling constants between ^{31}P–^{195}Pt spin pairs. We compare the results of GIPAW calculations with all-electron cluster-based calculations. We also evaluate the importance of relativistic effects by comparing calculations using the ZORA Hamiltonian at the scalar and spin-orbit levels. Finally, we contrast the results obtained through generalized gradient approximation functionals with those of hybrid functionals.

POSTER 218
Optimum Signal-to-Noise in Non-Uniform Weighted Sampling
Presenting Author: Len Mueller

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Non-uniform weighted sampling promises increased sensitivity while preserving the ability to generate the frequency domain spectrum through the application of a straightforward Fourier transformation. Yet there remains no consensus on how best to combine multiple samples at the indirect time points: should they be averaged or summed? The answer is neither. Here we demonstrate both theoretically and experimentally that the maximum signal-to-noise ratio for exponential NUWS is found when the signal is constructed using consistent RMS noise. This is in keeping with our earlier work that showed that NUWS UCR preserves spectral knowledge in both the time and frequency domains. Both analytic theory and experiment verify this conclusion.

POSTER 219
Acquisition of Ultra-Wideline NMR Spectra using Broadband Adiabatic-Inversion Cross Polarization: Applications to Quadrupolar Nuclei in Stationary Samples
Presenting Author: James J. Kimball

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Large anisotropic NMR interactions can give rise to ultra-wideline (UW) NMR powder patterns ranging from 250 kHz to upwards of 10 MHz in breadth, making it difficult to acquire uniform spectra with high signal-to-noise ratios. Frequency swept pulses have been shown to be superior to rectangular pulses for the acquisition of UWNMR powder patterns due to their large excitation and refocusing bandwidth capabilities. Additionally, cross polarization via the BRAIN-CP/WCPMG pulse sequence allows for broadbanded signal enhancements. Herein, we demonstrate the optimization and performance of BRAIN-CP/WCPMG for the acquisition of a series central transition UWNMR powder patterns of half-integer quadrupolar nuclei, which has hitherto been unexplored. The spin dynamics are investigated via numerical simulations and correlated with experimental results.

POSTER 220
NMRFAM User Program
Presenting Author: Paulo Falco Cobra

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The NMRFAM user program provides access to 11 NMR spectrometers (500 MHz – 900 MHz) equipped for a variety of solution and solid-state NMR experiments. Our scientists provide advice and assistance in experimental design, data acquisition and processing for studies of molecular structure, dynamics and interactions. We have experience with a range of sample types, including soluble and membrane proteins, fibrils, RNA, small molecules, and metabolomics. We have spectrometers equipped with 5 mm HFCN and HPCN QCI cryoprobes, and 1.7 mm HCN cryoprobe. We have multiple samplejet for automated data collection on small molecules and metabolomics samples. We have multiple 1.6mm (Varian) and 3.2mm (Varian or Bruker) MAS probes, both broadband and optimized for biological samples.

POSTER 221
National Resource for Advanced NMR Technology: Increasing NMR sensitivity to enable characterization of complex biomolecular systems
Presenting Author: Joanna Long

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The NIH-funded National Resource for Advanced NMR Technology focuses on the frontiers of NMR technology and their implementation for a broad user community pursuing fundamental questions at the forefront of biomedical research. Our first core features the use of high temperature superconductors (HTS) for RF coils, leading to high sensitivity for solution NMR spectroscopy. Our second core takes advantage of our 600 MHz MAS-DNP NMR instrument, which provides enhanced sensitivity through the transfer of magnetization from electrons to protons, and focuses on DNP probe development and sample preparation strategies specific to biomolecular samples. Our third core uses the 36 T Series Connected Hybrid magnet and the highest-field NMR spectrometer in the world for ssNMR characterization of quadrupolar nuclei.