Development of a Triple-resonance SAS Probe with Field Gradient Coil for Dynamic Control over Alignment of Proteins in Bicelles

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Long-range constraints from residual dipolar couplings in partially aligned solutions have been shown to be very useful in macromolecule structure determination. Partial macromolecule alignment has been obtained by using dilute liquid crystal solutions of disc-shaped bicelles, but this alone does not permit dynamic control over the alignment.

Switched Angle Spinning (SAS) techniques should provide the needed dynamic control over the bicelle (hence, the protein) alignment. When a sample containing discoidal bicelles of negative magnetic anisotropy is spun at 54.7^0 with respect to B_0 in Magic Angle Spinning (MAS), their interaction with B_0 vanishes and their orientation becomes random. For sample spinning at angles less than 54.7^0, they align with their normals perpendicular to the spinning axis, while spinning at greater angles causes their normals to align with the spinning axis. Dynamic control over the spinning axis is expected to provide the protein alignment control needed for more effective utilization of the bond angle information inherent in the residual dipolar coupling.

The effective protein methods require triple resonance and a PFG coil. Indirect (^{1}H) detection is also essential, so high sensitivity on the ^{1}H channel and high spectral resolution (0.005 ppm) are required. The proposed SAS techniques require rapid (~25 ms) reorientation of the spinning axis without adversely affecting spinning stability or rf tuning; and there are a number of additional requirements, including high-stability spinning speed detection, stable temperature control, low ^{1}H background signals, compatibility with narrow-bore (NB) high-field magnets, and possibly a ^{2}H coil for lock or decoupling.

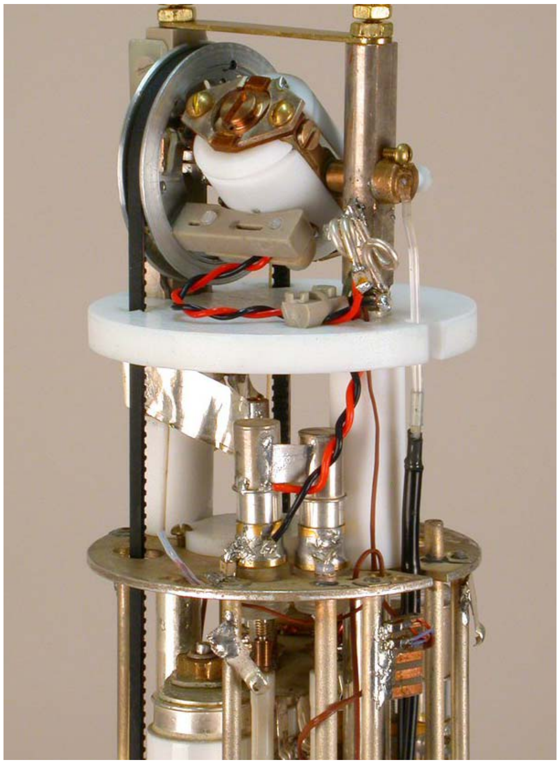

Figure 1. Photo of WB SAS probe with novel approaches to position encoding and rf leads. The commutator MF/LF lead connections are not visible in this view.

Two SAS probes suitable for such experiments in high-field magnets are under development – one for wide bore and one for narrow bore magnets, where some limitations in lead lifetime, PFG performance, and angle-setting accuracy appear likely. Novel approaches to the flexible rf lead problem and position encoding will be presented that have demonstrated ***lead life in excess of 300,000 flips and angular accuracy better than 0.1^o*** (a factor of four better than our prior methods) in the wide bore probe. The WB SAS probe head without the field gradient coil is shown in **Figure 1**. Preliminary experiments with a novel 4 mm spinner design compatible with automatic sample exchange have demonstrated 10 kHz spinning while flipping in ~25 ms. Achieving similar performance at 14 T required the development of a novel high-resistivity low-susceptibility alloy to minimize eddy currents during fast flipping.

Multi-coil rf circuits are shown to permit greatly improved sensitivity on all channels in the high-field triple-tuned SAS probe, where long leads are unavoidable. ***A low-E ^{1}H cross-coil achieves order-of-magnitude reduction in rf decoupler heating compared to multi-tuned solenoids***, and an outer solenoid is used for the mid-frequency (MF) and low-frequency (LF). Efficient H/X/Y performance is demonstrated with both 4 mm and 7 mm rotors.

Beta tests of the next generation of CST full-wave EM simulation software, MWS 2006, indicate it may finally have the long-sought-after ability to accurately and quickly simulate complex coils in which losses of all types (surface, dielectric, radiation, interactions, etc.) are significant and fine features are present in the geometry. RF coil optimization results and experimental validations will be presented, along with NMR test data. Preliminary tests on a 7 mm triple-resonance H/X/Y SAS probe with an actively shielded z-gradient coil for a 600 MHz wide-bore achieved very high efficiency on all three channels, with over 55 kHz ^{1}H decoupling at 120 W.

Reference:

G Zandomengehi, M Tomaselli, JD van Beek, and BH Meier, "Manipulation of the Director in Bicellar Mesophases by (Variable Angle) Sample Spinning: A New Tool for NMR Spectroscopy," *J. Am. Chem. Soc.,* 123, (2001) 910-913.