## Experimental Setup for DNP Spectroscopy and Variable Field Proton Electron Double Resonance Imaging

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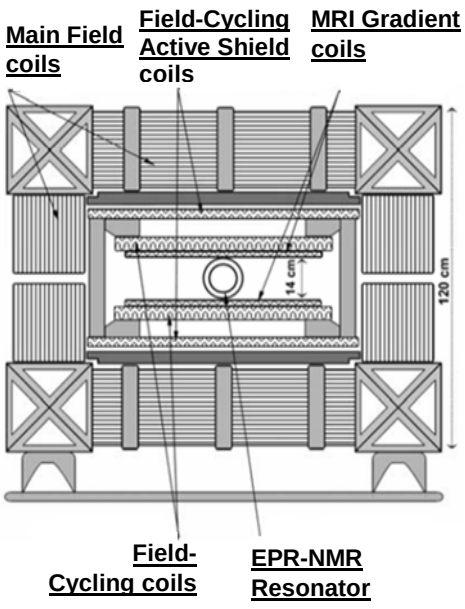
In the Overhauser effect, also known as dynamic nuclear polarization (DNP), the NMR signal of a paramagnetic solution is observed, typically the proton NMR signal of water, while sample is irradiated with the EPR resonance RF of the solute. Since proton-electron double resonance imaging (PEDRI) is based on the enhancement of the proton MRI image, it inherently offers high sensitivity, high spatial resolution and rapid image data collection. It circumvents the resolution limitations encountered in EPRI that occur due to the very broad linewidth of most paramagnetic labels. PEDRI is a very promising technique for in vivo imaging of free radicals in biomedical applications. DNP is useful for identifying free radicals and for studying EPR spectral characteristic that can provide important physiological information such as oxygen content or pH. The EPR spectroscopic information also contains a wealth of information about the environment of the radical label such as its mobility, macromolecular binding and localization in membrane versus aqueous environments. This information also enables study of the properties of mixtures of different paramagnetic species and allows these to be distinguished.

To further adapt the Overhauser effect for biomedical research, and to be able to perform field-cycled PEDRI at variable fields, a new system capable of performing DNP spectroscopy at variable NMR detection and EPR irradiation fields has been developed for small animal applications. A clinical MRI iron core magnet (Resonex Corporation) with vertically oriented magnetic field and a gap of 50 cm has been transformed for this purpose by the addition of a secondary electromagnet -- Field Cycling (FC) coils. The magnet is powered by modified Danfysik supply and allows working in current controlled mode at any field between 0.005 and 0.4 T, which allows utilizing frequencies up to 13 GHz for EPRI and 16.3 MHz for MRI. The magnetic field stability relies on the high precision manually adjustable reference voltage. Current regulation provides stability better than 0.5 ppm/hr of the current and hence magnetic field.

The FC coils (Tesla Engineering, West Sussex, UK) was built into the gap of the primary magnet and they can provide a magnetic field offset of up to 0.1 T to perform EPR irradiation at the low field followed by high field NMR detection. Its low inductance of 11 mH allows rapid current switching during acquisition of each point in the DNP spectrum. The FC coils power supply consists of a Copley 266 power supply amplifier (Copley Controls, MA, USA) rated at 350 V, 250 A which is powered by two 15 kW DC power supply (Lambda EMI, NJ, USA) connected in parallel. The output current of the Copley power supply amplifiers can be ramped up from 0-207 A in 8 ms which corresponds to a 0-670 G change in the vertical field produced by the cancellation coils. The FC coils are actively shielded to minimize the eddy currents that result in the primary electromagnet instability when the current in the FC coils is ramped. To preserve the original functionality of the clinical imager, FC coils were designed as a movable insert. The FC coils are mounted on a roller and rail mechanism to slide in and out of the magnet providing ease of interfacing and setup along with convenient access for service and further modifications of the Resonex scanner. The insert is also equipped with set of planar 3D field gradients and gradient power supply (Copley 235) 3 channel amplifier. This water cooled gradient set has a gap of 14 cm. Amplifiers are capable of 300 A per channel and can work in fast pulse mode for PMRI and also continuous for EPRI (35 G/cm along X, Y, or Z). Thus, the MR imager allows performing low-field MRI, high field MRI, EPR-NMR co-imaging, fixed-field PEDRI and variable field DNP spectroscopy with minimal time for switching between each modality.




**Mechanical layout of the variable field PEDRI/DNP spectroscopy system**

The system was equipped by a customized MRRS MR 5000 console (MR Research Systems, Surrey, UK). The console controls the gradient hardware, the RF system, field-cycling coils, EPR pulses, image acquisition and post-processing. The software comprises a digital signal processor, pulse sequence programmer, RF waveform generator and gradient waveform controller. A 12-bit MR3031 DAP converter was used to generate the necessary analog outputs required to drive the FC coils with 0.05 G resolution.

The performance of the system equipped with corresponding resonator sets was evaluated with nitroxide solutions filled phantoms. Variable field PEDRI and DNP modalities at detection fields of 97 G, 200 G and 587 G were employed. We believe that this instrument will be useful in the application of PEDRI techniques for in-vivo studies and will be indispensable for optimization of the EPR excitation and NMR detection parameters for given biomedical applications.

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