Nuclear magnetic resonance (NMR) is a physical phenomenon in which the magnetic moments of atomic nuclei interact with an external magnetic field. By utilizing this interaction we can study the structure and dynamics of molecules in of sizes ranging from small organic compounds (like ethanol or formic acid) up to biological macromolecules such as proteins and nucleic acids (the molecules carrying genetic information). NMR experimental techniques are a important tool used to study the molecular basis of biological processes, including diseases. Of great importance is its ability to study both local and global structure of proteins, as its elucidation is a major step towards understanding their function and in case of disease-related proteins also rational drug design. Together with X-ray diffraction NMR is one of the two major methods allowing for structural determination with atomic resolution and the only one capable of performing this task in conditions similar to these in living cells.

The main drawback of magnetic resonance studies is poor experimental sensitivity, which largely determines experiment times. This is especially problematic owing to the high cost of NMR spectrometers and non-trivial production of proteins samples. Methods for detailed structural studies of smaller proteins and the determination of global shape of even very large proteins (or their complexes with other proteins and/or other molecules) are well developed. What causes this is the fact that with increasing molecular mass the interactions between magnetic moments within the molecule become so strong, that the signals of interest in NMR experiments will strongly decay before we can measure them. This issue can be sidestepped by substituting most of the protons (hydrogen nuclei) in a molecule with another isotope of hydrogen - deuterium (composed of a proton and a neutron, its nucleus is called a deuteron) with has far weaker magnetic properties. For large proteins this leads to a dramatic increase in the intensity of signals registered from the non-substituted protons, but with only a fraction of possible proton positions left (generally these that can exchange with water protons) the resolution of obtained structural information is considerably lower. By selectively inserting protons in well-defined position in molecules (by growing the protein-producing bacteria on selectively labelled compounds) this problem can be ameliorated but such methods are both costly and laborious.

The aim of this project is to develop new experimental methods (sequences of magnetic field pulses) that would enable the collection of high-resolution structural information on large proteins by utilising randomly fractionally deuterated samples. Such samples are prepared by using a mix of protonated and deuterated compounds and results in proteins in which each proton position is substituted randomly by deuterium in for example 70% of molecules. Although the maximum signal intensity for such a position will be reduced (here to 30%), the smaller total number of protons in individual samples will partially compensate for this loss. This effect alone is not enough to raise the sensitivity of the experiments substantially. In the methods we intend to develop we will make use of other effect. Foremost of these is the possibility of eliminating the main source of signal attenuation - dipolar interactions between protons and the nuclei of atoms they are bound to - carbons and nitrogens. The elimination of this effect by employing the so-called multiple-quantum coherences has limited utility in non-deuterated proteins due to the destructive influence of neighbouring protons on such coherences - but the number of such protons in randomly fractionally deuterated proteins is considerably lower! The utilization of this phenomenon together with other experimental optimizations should allow for collection of high quality structural information for proteins that are too large for detailed characterization using current NMR methods and do not form crystals suitable for X-ray diffraction.