Abstract

This thesis describes detailed NMR investigations of the structural and dynamic characterization of biomolecules such as peptides, proteins, carbon nanotubes and nanoparticles. Specifically, the thesis focuses on the determination of major secondary structures of proteins from six backbone nuclei NMR chemical shift values, the determination of conformational dynamics of novel tripeptides using NMR experiments and quantum chemical computations, the distinction of different types of single-walled carbon nanotubes using 19F NMR shielding tensor, characterization of protein-nanoparticles interaction using NMR, protein-ligand binding identification using 19F NMR diffusion and relaxation, and the tomography of 19F shielding tensor of fluorinated tryptophan using cross-correlated spin relaxation. Chapter 1 This chapter provides a basic introduction to the theory of NMR spectroscopy, a brief description of the NMR signal and spectrometer hardware, the chemical shielding (CS) tensor, computational chemistry to find the CS tensor of NMR active nuclei and a background to the NMR studies of nanoparticles interacting with biomolecules. Chapter 2 This chapter describes a method we developed called 3DChemCorr, based on the distinction of three major secondary structure helix, sheet and turn of protein using three-dimensional NMR chemical shift correlation maps of different combinations of six backbone nuclei namely 1HN, 15N, 13Cα, 1Hα, 13Cβ and 13C ′. NMR chemical shift values of six backbone nuclei of alanine, proline and cysteine amino acids of 579 protein from PDB database were extracted using python programs and their secondary structure were determined using four different secondary structure determining programs: MOLMOL, STRIDE, VADAR and DSSP. Two and three dimensional chemical shift correlation maps of different combinations of six backbone nuclei were plotted and probabilities were determined using Monte-Carlo simulationsfor three major secondary structures: helix, sheet and turn. The three-dimensional correlation map was found better in distinguishing turn secondary structure as compared to two-dimensional maps. We employed this method on six different proteins including two membrane proteins and compared our result with the already existing methods of secondary structure determination using NMR chemical shift values. Our method was better at predicting the turn secondary structure as compared to standard predictive methods and comparable to standard methods whilst predicting helices and sheets. Chapter 3 This chapter deals with the determination of conformation of the novel tripeptide arg-glu-arg (and its analogues constructed using D-forms of aminoacids) using liquidstate NMR experiments and quantum chemistry calculations performed using Gaussian09 package. The four different regions alpha-helix, beta-strand, ppII and left handed alpha helix of Ramachandran map were selected for computation and stable conformation was determined, which was then compared with liquid-state NMR experiments. Chapter 4 This chapter deals with the characterization of the interactions of gold and silver nanoparticles with proteins lysozyme and cytochrome c using 1H-13C NMR chemical shift correlation experiments. Chapter 5 This chapter describes the characterization of fluorinated single-walled carbon nanotubes using the 19F chemical shielding (CS) tensor computed using Gaussian03 quantum chemistry software package. Our theoretical investigation is based on the comparison of CS tensors of 19F nuclei of zigzag and chiral single-walled carbon nanotubes with different 19F substitutions and multi-dimensional correlation plots of different CS parameters. 19F MAS powder pattern spectra were simulated using the SIMPSON package for solid-state experiments to perceive the differences. Furthermore, 19F CSA-DD cross-correlated relaxation rates for liquid-state NMR experiments are also predicted. All these methods are found to be able to clearly distinguish between the two forms of single-walled carbon nanotubes (chiral and zigzag). Chapter 6 This chapter is based on the identification of the competitive binding of three fluorinated ligands namely 5-fluoro-tryptophan, 4-fluoro-phenylglycine and 5-fluoro-uracil to bovine serum albumin (BSA) protein using 19F NMR experiments. The evidence of binding of 5-fluoro-tryptophan out of all three ligands was confirmed using onedimensional 19F NMR, 19F NMR diffusion experiments, 19F T1, T2 relaxation experiments and 19F-1H HOESY experiments. Chapter 7 This chapter describes the experimental and computational methods used to fully characterize the fluorine CS tensor in a fluorinated aminoacid 5-fluoro-tryptophan. The experimental chemical shift anisotropy-dipolar (CSA-DD) relaxation rates of fluorinated tryptophan were determined

using a set of cross-correlated spin relaxation liquidstate NMR experiments. The result was compared with quantum chemistry calculations performed using Gaussian03 package and previously determined solid state NMR experimental results. Chapter 8 A brief outline of the main results of the thesis are summarized and some prospects for future extensions of the work are described in this chapter.