Synopsis

Chapter 1. Nanomaterials have been used by mankind for centuries without knowing the dimensions of the materials they used. However, it is only the last ten years or so that the subject of nanoscience and technology has gained tremendous attention. From the biological and medical applications viewpoint, the primary interest in nanoparticles stems from the fact that they are small enough to interact with biomolecule (protein, DNA, carbohydrate). Interestingly, the interaction between nanomaterials and bio-molecules results in the formation of a biological corona on the NP’s surface that is quite dramatically different from that adsorbed on a flat surface of the same bulk material. In the present chapter a brief description has been made about nanoparticle, its characterization, and interaction with protein. This knowledge is important from the perspective of safe use of nano materials.

Chapter 2. Nanoparticles (NP) have wide range of applications in the field of medicine and thus contributing to one of the most emerging areas of medical research. Once nanoparticles are exposed to biofluids they get coated with proteins. As proteins are adsorbed on the surface, the extent of adsorption and the effect on the protein conformation and activity are dependent on the chemical nature, shape and size of the nanoparticle. In this chapter, we have investigated the interaction between ZnO nanoparticles (NPs) with structurally diverse set of proteins, such as lysozyme, α-lactalbumin and bovine serum albumin (BSA) using calorimetric and other biophysical techniques. The results are interpreted in terms of the three-dimensional structure. Finally the results were compared to get a better insight into the process of protein nanoparticle interaction. The present study thus provides useful insights into issues such as protein-protein and protein-nanoparticle recognition.

Chapter 3. Zinc oxide nanoparticles (ZnO-NP) were synthesized by alcoholic route using zinc acetate as precursor material and lithium hydroxide as hydrolyzing agent. Further ZnO-PEI (ZnO derivatives) was made in aqueous medium using capping agent polyethyleneimine (PEI). X-Ray Diffraction (XRD) measurement confirmed the
formation of ZnO NP with hexagonal wurtzite structure. The average size of PEI functionalized ZnO (3-7 nm) was determined by using Scherer's equation and confirmed by transmission electron microscope (TEM). ZnO-PEI is water soluble and forms colloidal suspension in water. These functionalized nanoparticles along with uncapped ZnO showed promising antibacterial activity against a range of multidrug resistant bacterial species. ZnO effectively killed these microorganisms by generating reactive oxygen species (ROS). A synergistic effect was further seen when nanoparticle was co-treated with tetracycline.

Chapter 4. The antibacterial effect of polyethylene imine (PEI) functionalized zinc oxide (ZnO) nanoparticles on Helicobacter pylori was investigated for inhibition and inactivation of cell growth. The results showed that H. pylori was extremely sensitive to treatment with PEI-capped ZnO nanoparticles. Internalization and uniform distribution of PEI-capped ZnO without agglomeration was observed in H. pylori cytosol by electron microscopy and Energy dispersive x-ray measurement (Edax). The MIC of ZnO nanoparticles for H. Pylori was determined to be 0.01 mg/ml. Scanning electron microscopy examination revealed that the majority of the cells transformed from spiral shapes into coccoid forms after exposure to 0.02 mg/ml of ZnO nanoparticles for 1 h, which is consistent with the morphological changes of H. Pylori under other stress conditions. These coccoid cells were found to have a certain level of membrane leakage. To address the molecular basis of ZnO nanoparticle action, a large set of genes involved in cell stress response, virulence, and housekeeping were selected for gene expression study. Reverse transcription-quantitative PCR (RT-qPCR) showed that in response to treatment with ZnO nanoparticles, the expression levels 16s and 23s RNA were worse affected. A further confocal microscopy investigation revealed that ZnO can induce DNA damage through production of reactive oxygen species (ROS). These results suggest that the antibacterial mechanism of ZnO nanoparticles is most likely due to generation of reactive oxygen species and disruption of housekeeping gene expression. Our investigations clearly exhibit the potential of ZnO-PEI nanoparticles as an alternative for conventional gastric ulcer treatment.
Chapter 5. Nanoparticles are increasingly recognized for their utility in biological applications including nanomedicine. The present study investigated the toxicity of zinc oxide (ZnO) nanoparticles toward highly proliferating breast cancer cell such as MCF-7, MDA-MB etc along with normal cells like peripheral blood mononuclear cells (PBMCs). In this investigation, we explored the mechanism by which ZnO nanoparticle induces apoptosis in different breast cancer cells. It was observed that Fas-FADD pathway was activated in breast cancer cells upon ZnO nanoparticles treatment. Fas-FADD is one of the crucial apoptosis pathways by which external ligands induce apoptosis. Fas-FADD activation ultimately resulted in cleavage of BID and alteration of mitochondrial transmembrane potential that leads to cytochrome-c release and activation of several caspases. Although this is the major pathway involved in cancer cell apoptosis, possibility of ROS mediated intrinsic apoptosis pathway could not be ruled out.

Chapter 6. Curcumin is known for its anti-carcinogenic properties, although the exact mechanism of its action or the identity of the target receptor is not completely understood. Studies on a series of curcumin analogs, synthesized to investigate their tubulin binding affinities and tubulin self-assembly inhibition, showed that: i) curcumin acts as a bifunctional ligand, ii) analogs with substitution at the diketone and acetylation of the terminal phenolic groups of curcumin are less effective, iii) a benzylidene derivative compound (7) is more effective than curcumin in inhibiting tubulin self-assembly. Cell-based studies also showed (7) to be more effective than curcumin. Using fluorescence spectroscopy we show that curcumin binds tubulin 32 Å away from the colchicine-binding site. Docking studies also suggests that the curcumin-binding site to be close to the vinblastine-binding site. Structure activity studies suggest that the tridented nature of compound (7) is responsible for its higher affinity for tubulin compared to curcumin.