# Prologue

I was first introduced to the field of Enzymology and Enzyme Technology when I joined as a Ph.D. student in July 2001, by my supervisor Dr. Arvind M. Kayastha and my Ph.D. work is a potpourri of studies using the enzyme urease from a plant source. Since urease is a classical enzyme, and hence an extensive review has been presented in the **Chapter 1** of this thesis.

*"Inclusion bodies are major bottlenecks in protein production and are hampering the development of top priority research areas such as structural genomics"*..... this very statement has sparked my interest to investigate the conditions for renaturation of denatured urease from jack bean (**Chapter 2**). This work was initiated in School of Biotechnology in collaboration with Molecular Biology Unit, Banaras Hindu University and could be completed when I had an opportunity to visit Institute for Biotechnology, Martin-Luther-University Halle-Wittenberg.

*"When the single use of enzyme is economically not feasible, the enzymes can be reused"*; the immobilized enzymes are becoming increasingly popular as reusable, selective analytical chemical reagents in solid-phase flow-through reactors, as membranes in sensors and as films in dry reagent kits (**Chapter 3**). A simple to use, inexpensive, reliable, efficient and flexible method of immobilization for urease using DEAE-Cellulose paper (dipstick kit) was developed. This has been discussed in **Section 1, Chapter 3**.

*"Apart from cost, the limiting factor for immobilization technology is the stability of enzyme"*; immobilization of urease on Alkylamine and Arylamine

glass resulted in very high immobilization and improved stability. This aspect has been discussed in **Section 2, Chapter 3**. An attempt was made to measure the urea concentration in serum samples using potentiometric method with glass-immobilized urease.

"*The need for biosensors with a high degree of accuracy and specificity is critical for a range of applications*", from biomedical monitoring to sensors for security and biohazard detection. To this end, a novel fabrication was developed and this single biosensor comprised of two detection systems (amperometry and potentiometry), wherein two different signals are measured simultaneously for a single analyte (**Chapter 4**). This work was carried out at Electro-analytical & Sensors Lab, Ruhr-University Bochum.

"*Apart from agricultural and pharmaceutical importance*", studies on urease inhibition would also provide insight into the mechanism of catalysis (**Chapter 5**). This chapter deals with inhibitions studies on boric and boronic acids **(Section 1)**. Furthermore, hexameric pigeonpea urease demonstrated molecular asymmetry with pH inactivation **(Section 2)**.

This Ph.D. dissertation has resulted in five publications (list attached in the end of the thesis).