Summary

Different types of pathogenic bacteria cause food poisoning. In most cases, food poisoning are caused by *Staphylococcus aureus, Salmonella sp, Clostridium perfringens, Campylobacter sp, Listeria monocytogenes., Vibrio parahaemolyticus, Bacillus cereus,* and entero-pathogenic *Escherichia coli* which produce a number of toxins. These bacteria are commonly found in raw foods. Since food-pathogenic bacteria are often present in many foods, knowing the characteristics of such bacteria is essential for effective diagnosis and control.

The commonality in microbial virulence mechanisms and the occurrences of similar resistance systems in animals and plants point out that all these mechanisms have an ancient and intertwined history. It is quite evident that susceptibility or resistance to disease involves subtle and highly specific exchanges of molecular signals between pathogens and their hosts and a clear-cut understanding of these mechanisms can provide newer approaches to diagnose and control diseases. The genomic islands and operons are considered as the units where groups of genes are transcribed together and whose products contribute to specific function. One of the typical examples of genomic islands is the pathogenic island (PAIs), which is present in pathogenic bacteria that form the principal molecular component responsible for the development of a specific disease. Codon usage study provides information of use of different codon in a genome, as it is often seen that all codons are not used evenly. A detailed and accurate analysis of codon usage is an essential prerequisite to our understanding of how and why divergent patterns of codon choice evolved. Genomics has a great potential in the study of food pathogens showed the relationship between the predicted level of gene expression based on codon usage, actual microarray expression values and gene function at the genomic level in *S. pneumonie.*

Although a lot of work has been performed and is going on in codon usage of different microorganisms but very little work performed on codon usage of food pathogenic bacteria. To understand the mode of action of toxin it is very much necessary to known their three dimensional structure and motional properties. Crystal structures of several toxins have been solved which enlighten us about their structure. But the structures of many toxins are yet to be solved. Molecular modeling &molecular dynamics of some toxin have been done. In this work firstly we would try to characterize the pathogenicity island and toxic genes by statistical analysis, secondly knowledge based model will be built to get three-dimensional structure of toxins. This thesis contains total eight chapters. First and Second chapter describe introduction and review of this work. Third chapter describe codon usage patterns of five complete genomes of *Salmonella*, predict highly expressed genes, examine horizontally transferred pathogenicity-related genes to detect their presence in the strains, and scrutinize the nature of highly expressed genes to infer upon their lifestyle where Protein coding genes, ribosomal protein genes, and pathogenicity-related genes were analysed with Codon W and CAI (codon adaptation index) Calculator. Fourth Chapter describe Bioinformatic study of Pathogenicity related genes of three species of Helicobactor where Protein coding genes, ribosomal protein genes, and pathogenicity-related genes were analysed with Codon W and E-CAI (codon adaptation index) server. In the fifth chapter describe Characterization of pathogenic genes through condensed matrix method, case study through bacterial zeta toxin. In this study, zeta toxin nucleotide sequences of some pathogenic and non-pathogenic Bacteria were used for phylogenetic analysis. The uniqueness of this method is that it does not employ sequence alignment of complete nucleotide sequence of the corresponding gene.

In the chapter six describe Molecular Dynamics Simulation Receptor-Binding C-Terminal Domain from *Clostridium difficile* Toxin A to understand the motional properties and mode

of action of the receptor-binding C-terminal domain of *C. difficile*, molecular dynamics simulation of *C. difficile* toxin A in aqueous environment was carried out using the software, GROMACS. From the time evolution, Root Mean Square Deviation (RMSD), Root Mean Square Fluctuation (RMSF) and Radius of gyration (Rg), it was found that the toxin was relatively inflexible. Principal Component Analysis (PCA) was also performed for better understanding of motional properties in reduced dimension. Analysis of binding site reveals that Ala51, Ala58, Ile59 and Tyr93 have very low fluctuation. All these observations help us to understand the mechanism of pathogenesis related with toxin A of *C. difficile.*

In the chapter seven describe Comparison in motional properties of *Staphylococcus aureus* exfoliative toxins A and B as revealed by their MD simulation to understand the motional properties and mode of action of *Staphylococcus aureus* exfoliative toxins A and B, molecular dynamics simulation are carried out using the software GROMACS. From the time evolution RMSD, RMSF and Radius of gyration, it is found that the toxin A and B are not much flexible and it is also indicated by principal component analysis. Pro192 and val183 is key residue towards the activity of toxin A and toxin B respectively. It is seen dihedral angle psi of Pro192 of toxin A is free to rotate without involvement of much energy and its becomes active after a conformational triggering of this dihedral but Val 183, the corresponding residue of toxin B, shows very less conformational freedom.

In the last chapter describe Homology modeling and MD simulation of the CdtB of *Helicobacter hepaticus* ATCC 51449 where the 3D model of the CdtB of *H. hepaticus*ATCC is constructed by MODELLER 9v4 program using the templates CdtB from *Actinobacillus actinomycetemcomitans.* The model is validated by PROCHECK, ProSa, CASTp server, ProFunc server etc. After that molecular dynamics simulation is performed using GROMACS and the resulting trajectory is analyzed. Homology modeling

can produce high-quality structural models when the target and template are closely related, which has inspired the formation of a structural genomics consortium dedicated to the production of representative experimental structures for all classes of protein folds (Williamson AR, 2000). Like other methods of structure prediction, current practice in homology modeling is assessed in a biannual large-scale experiment known as the Critical Assessment of Techniques for Protein Structure Prediction, or CASP.

The wealth of information obtained from the genome projects needs to be mined. As newer and newer toxins genes are discovered and sequenced novel insights are being gained. Bioinformatics in combination with metagenomics as well as metaproteomics approaches has the potential to give a more detailed picture that underlies pathogenicity as well as diseases. In absence of crysllographic or NMR structure Homology modeling will enlighten us about three dimensional structure and molecular dynamics simulation opening newer possibilities for exploring the molecular mechanism and activity of that toxin. At the end of the day scientific perception will continue to play a vital role in creating models that clarify the functions of pathogens in improved manner.