CHAPTER 5

Summary
Helicobacter pylori, a Gram negative curved bacterium has been identified and related to the gastro duodenal pathologies in 1984 AD by two Australian scientist Dr. Barry Marshal and Dr. Robin Warren and since than lot of work has been done on this bacterium, two of the H. pylori strains ATCC 26695 and J99 has been completely sequenced and compared, this has began a new era in H. pylori research.

Since H. pylori is a fastidious bacterium and requires special techniques and expertise for it’s culturing such as microaerophilic conditions, Horse or sheep blood and other sophisticated techniques, it is not possible to culture H. pylori in the limited resources and hence it was necessary to find out the alternate source for the microaerophilic conditions and to develop the cost effective techniques for its isolation. H. pylori culturing technique using the candle jar method which was proved to be the cheapest and the easiest method of H. pylori isolation has been established, more over isolation of H. pylori in candle jar desiccators is a more liable and flexible method, which gives more than 80% of accuracy and also that this is the cheapest method for the culturing of H. pylori, as far as developing countries like India is concern, the combination of Urease test, Culture and Histopathology combining together will give more accuracy than the individual test along with the conformation by polymerase chain reaction.
Helicobacter pylori strains isolated from the Indian patients have been genotyped for the plasticity region ORFs and it has been found that the isolated strains has more or less intact plasticity region in the representative disease categories, some of the ORFs such as JHP0947 and JHP0940 has been found to be associated with Duodenal ulcers and Gastric carcinoma respectively, however this should be tested against many more isolates of *H. pylori* from different geographical regions and from different disease categories to arrive to an exact conclusion.

The plasticity region ORFs JHP0940 and HP0986 has been expressed in *E. coli* and the recombinant protein was purified in soluble form using an on column folding strategy, this has also suggested that these ORFs are not hypothetical proteins.

The gene product of the hypothetical JHP0940 and HP0986 protein codes for an alpha helical protein as confirmed by the results of the in silico analysis using predict protein PSIPRED and DNASTAR protein analysis softwares.

The protein JHP0940 is not stable at room temperature and also at 4, -20, and -80° C as compare to the other recombinant protein *i. e.* HP0986 which is stable at 4 °C and less temperature.

HP0986 elicits a strong immune response in all the four clinical categories of patients included in this study.

A strong immune response to HP0986 was observed in the *H. pylori* cases as compared to ICD protein, whereas the reactivity to ICD and HP0986 in all the four categories of the patients was comparable, pointing to the immunodominant nature of this protein.
Recombinant protein HP0986 distinguishes between the *H. pylori* infected and not infected persons. The antibody response to HP0986 is same as that elicited by ICD in patients infected with *Campylobacter jejuni*, both the antigen showed almost equal reactivity towards the *C. jejuni* patients as compare to the healthy controls, this can easily rules out any possibility of cross-reactivity.

HP0986 can be used as a possible diagnostic marker for *H. pylori* infected people. The immune response to HP0986 in the case of healthy controls and *C. jejuni* was comparable. HP0986 however mounted statistically significant immune response to sera obtained from *H. pylori* infected patients including all the four infected categories, pointing to the ability of this ORF to differentiate between *H. pylori* infected and not infected persons.

In conclusion, data presented in this study describe for the first time, a complete picture of the prevalence of the plasticity region ORFs in the *Helicobacter pylori* strains isolated from Indian patients, immunodominant as well as structural characteristics of a hypothetical protein belonging to the plasticity region of *Helicobacter pylori*.

In view of such findings it can be speculated that some members of the plasticity region cluster provide selective advantage to some of the strains to adapt to changing host niches and become more and more invasive. In what way such advantage is gained? This needs to be discovered.