Detection of Non-pylori Helicobacter Species DNA in Human Gastroduodenal Biopsies by a 23S rRNA Gene-based PCR-RFLP Methods

Abbas Yadegar; Masoud Alebouyeh*; Tabassom Mirzaiee; Ehsan Nazemalhosseini Mojarad; Mohammad Reza Zali

Research Center for Gastroenterology and Liver Diseases, Shahid Beheshti University of Medical Sciences, Tehran, Iran
babak_y1983@yahoo.com

Background & Objectives: Besides Helicobacter pylori, other Helicobacter species have also been associated with gastritis, peptic ulcers, gastric cancer and mucosa associated lymphoid tissue (MALT) lymphoma in humans. Non-pylori Helicobacter infections of the human stomach are mostly accompanied by active chronic gastritis. However, gastric erosions located mainly in the antrum and duodenal ulcers have also been reported in association with non-pylori Helicobacter infections. The main goal of this study is to investigate presence of non-pylori *Helicobacter spp*. DNA in human gastroduodenal biopsies.

Methods: A total of 107 patients suffering from gastroduodenal diseases, whom underwent endoscopy during 2010-2011 in Tehran, was included in this study. Gastric and duodenal biopsy samples were taken from all patients and kept at -70° C until used for DNA purification and PCR. Two Helicobacter genus-specific PCR assays using 16S and 23S rRNA specific primers were performed on all of the DNA samples purified from biopsies. For detecting *H. pylori* DNA in the biopsies, the presence of ureC fragment (glmM gene) was also identified by PCR. To distinguish between Helicobacter species, RFLP analysis was carried out on all 23S rRNA fragments using double digestion with SmaI and HindIII endonucleases. In addition, 16S rRNA sequencing was carried out on all positive samples.

Results: Helicobacter genus-specific 16S rRNA primers identified the presence of Helicobacter DNA in 26 (24.3 %) of 107 patients. These positive samples were further confirmed as *H. pylori* by species-specific PCR, 23S rRNA RFLP analysis and 16S rRNA sequencing. Moreover, no DNA samples associated to non-pylori Helicobacter species was identified among the tested samples.

Conclusion: In spite of recent reports from western countries showing an association between gastroduodenal diseases and non-pylori Helicobacter infections, it is difficult to infer this relationship in Iranian population regarding our findings. It is likely that most non-pylori Helicobacter infections are acquired from direct contacts with animals. Therefore, further studies on patients keeping pets may give us more insights on the putative role of these organisms in human diseases.

Keywords: Non-pylori Helicobacters; *H.pylori*; 23S RRNA Gene; PCR-RFLP