**Title: GENOTYPIC ANALYSIS OF *Helicobacter pylori* BY MULTIPLE-LOCUS VARIABLE-NUMBER TANDEM-REPEATS METHOD IN SOUTHERN BRAZIL**

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**Abstract:**

*Helicobacter pylori is* a bacteria that infects about 50% of the world's population and is considered a major risk factor for the development of gastric diseases, including lymphoma and gastric cancer. Molecular tools based on the technique of PCR have been developed in order to characterize genotypically prokaryotes and eukaryotes. Despite to be used as a genotyping method for different microorganisms, few studies have reported the use of Multiple-Locus Variable Numbers of Tandem Repeats Analysis (MLVA) for analyzing the clonal diversity of *H. pylori*. The aim of this study was to analyze the genetic variability of *H. pylori* strains by MLVA method in ninety five DNA samples obtained from gastric biopsy of *H. pylori*-positive patients originating from two cities in southern Brazil. Initially, an *in silico* analysis of the 12 *loci* was performed in order to confirm the repeat sequences and the annealing sites of the primers previously described. The *in vitro* analysis was performed by MLVA method, using PCR and agarose gel, for *H. pylori* genotyping. From the 12 *loci*, four were viable for genotypic analysis. Five showed identity to more than one genomic region, which would lead to non-specific amplifications; one was not possible to standardize; and two resulted in fragments with similar sizes, indistinguishable by agarose gel. From the results obtained, ninety strains were distributed in thirteen different patterns, five strains had unique profiles and samples from Pelotas and Rio Grande shared similar genotypes. This can also be explained due to geographical proximity and ease of clonal dispersion of strains between the cities and the low number of viable *loci* for genotypic analysis. These *loci* showed different discriminatory indexes (HGDI values ranging between 0 and 0.67) and our findings reinforce the need to identify new markers in strains of *H. pylori*, to obtain a better discrimination of clonal diversity and greater accuracy of the method employed.

**Key-words:** *Helicobacter pylori*, genotyping, MLVA.

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