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幽門桿菌特殊之重組酵素(recombinase)之發現與功能分析
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幽門桿菌特殊之重組酵素(recombinase)之發現與功能分析

Isolation and characterization of novel recombinase(s) from *Helicobacter pylori*

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To identify genes that are involved in DNA recombination, we have screened 1500 mutant strains of a *Helicobacter pylori* mutant library. (1) One of the mutants showed severely impaired natural transformation by using chromosomal DNA as DNA donors. The inverse polymerase chain reaction and DNA sequencing revealed that the interrupted locus of the mutant was HP1473. The immediate open-reading frame downstream of HP1473 was HP1472. The knockout of HP1472 did not affect the frequency of natural transformation. The deletions of HP1473 in the NTUH-C1 and 26695 strains both impaired the ability of natural transformation. The complementation of HP1473 to the mutant partially restored natural competence. DNA binding and uptake assays revealed that the mutant was impaired in both. However, the HP1473 knockout mutant could be transformed with shuttle vectors but not with other homologous DNA donors by electroporation. The recombinant HP1473 protein was able to promote the formation of stable DNA homologous recombination on a nitrocellulose membrane and had DNA binding activity. The HP1473 knockout mutants also showed increased susceptibility to metronidazole and UV lights. These results indicate that HP1473 is involved in DNA binding/uptake and

recombination in *H. pylori*. (2) Another mutant showed severely impaired natural transformation by using chromosomal DNA and shuttle vectors as DNA donors. This mutant also showed decreased adhesion to two gastric epithelial cell lines. Inverse polymerase chain reaction revealed that the interrupted locus of this mutant was a HP0015 homologue of *H. pylori* strain 26695. DNA sequencing and reverse-transcription PCR revealed that HP0015 and 2 downstream genes (HP0016 and HP0017) were a transcriptional unit. Deletion and complementation constructs revealed that HP0016 and HP0017 were also involved in natural transformation, but not in adhesion. *Hp0015* was associated with both adhesion and natural transformation. Co-culture of a wild-type strain with AGS cells revealed that 19 genes in AGS cells were upregulated; however, five of the 19 genes were not induced by co-culture with *hp0015* mutants. These results indicate that *hp0015* is associated with adhesion, and *hp0015*, *hp0016*, and *hp0017* are associated with natural transformation.