PHOB/PHOR OF VIBRIO CHOLERAE O1*:* CHARACTERIZATION OF THE PROMOTER REGION AND IDENTIFICATION OF NEW PUTATIVE PHO REGULON GENES

Diniz MMP; Ladeira TC; Goulart CL; Barbosa LC; Pacheco ABF; Bisch PM; von Krüger WMA

Unidade Genômica, IBCCF, UFRJ, Rio de Janeiro, Brazil

PhoB/PhoR system regulates gene expression in response to inorganic phosphate (Pi) limitation in bacteria. PhoB, the response regulator, binds one or more 18bp sequences (*pho* boxes) upstream the Pho regulon genes to activate/inhibit transcription. In previous work we showed that mutation in *V. cholerae phoB* affected the Pi-starvation response and intestinal colonization. In the present work we describe the characterization of the *phoB/phoR* promoter region and new putative PhoB/PhoR regulated genes in *V. cholerae* O1. Analysis of 200 bp upstream *phoBR*, using ClustalW®, identified 5 putative *pho* boxes, on both, sense (3 boxes) and antisense (2 boxes) strands. Footprinting assay showed that PhoB binds boxes at: -35 (box1), -60 (box2) and -80 (box3). Functional studies using reporter gene fusions (*phoBR* promoter fragments:*lacZ*) showed that PhoB~box1 induces expression of *phoBR* and PhoB~box2 reduces the inducibility of *phoBR* by PhoB~box1. Four putative Pho regulon genes have been investigated: *aphB* (for a virulence regulator), *iciA* (for a DNA replication initiation inhibitor), *surE* (for a protein required for stationary-phase survival) and *phoE* (for a phosphorin). *pho* boxes, apparently active, were detected on their promoters: they all bind PhoB and induce expression of the corresponding genes. These results suggest that PhoB/PhoR system regulates a broad range of functions, including virulence, in *V. cholerae* O1.