

Polymerase chain reaction and cloning of *Burkholderia pseudomallei* putative genes.

ABSTRACT

Total of 23 putative Open Read Farms from *B. pseudomallei* strain D286 was successfully cloned and the nucleotide sequence analysis of the putative genes showed the homologue (98-100%) to strain K96243. The high similarity in gene sequences between these strains is confirmed for presence of the necessary ORF for LPS biosynthesis through PCR amplification the application of the ORFs in the PCR amplification and expression method. The findings of this study have contributed to some information on the molecular bases of the LPS biosynthesis genes in *B. pseudomallei* specifically for strain D286. PCR amplification, a specific pair of primer for each ORFs was proving specific for amplification of genes in *B. pseudomallei* strain D286. The PCR mixture with addition of DMSO, formamide and glycerol could ease the PCR optimization where different pairs of primers were involved. The specific primer pairs with the PCR mixture could be used in developing a PCR diagnosis of melioidosis.

Keyword: Polymerase chain reaction; Cloning; Melioidosis; *Burkholderia pseudomallei*; Open reading frames; Putative genes; Primers.