
Temperature-dependent expression of virulence genes in *Vibrio parahaemolyticus* AHPND strain (Vp_{AHPND})

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Abstract The risk of acute hepatopancreatic necrosis disease (AHPND) outbreaks in the shrimp farms are governed by interactions between host, pathogen, and environmental factors. Among the environmental factors, temperature plays distinct role in triggering diseases. Temperature fluctuation is not only an acceleration–deceleration modulation of the whole cell’s enzymatic activity but also affects membrane-associated functions and leads to changes in bacterial virulence gene expression. Moreover, temperature is one of abiotic stresses which arouse pathogen infection by changing host susceptibility. In the present study, we investigated the effect of different diurnal temperature fluctuation on the growth and expression of virulence genes of *Vibrio parahaemolyticus* AHPND strain (Vp_{AHPND}) cultured in a bioreactor. The pathogen, which had evolved under 3 different temperature patterns for 4 days, and an ancestor of the evolved strains (AAHMRU04), were observed for their virulence gene expressions by using quantitative RT-PCR. The mRNA expression levels for 3 virulence factors including Photorhabdus insect-related (Pir) A toxin and Pir B toxin and membrane-associated transcriptional factor (ToxR) were measured. The transcriptional analysis demonstrated that all evolved strains exhibited the expression levels of virulence genes different from those of the ancestor, which was further supporting its pathogenicity. In conclusion, the temperature fluctuation is an important environmental factor that influences the virulence of Vp_{AHPND} and these results would contribute to further study on the pathogenesis and host defensive system in the diurnal temperature fluctuation.

Keywords: Acute hepatopancreatic necrosis disease, Temperature, Virulence gene expression, *Vibrio parahaemolyticus*

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