Different isolates of *Bacillus subtilis* from gastrointestinal tract of wild caught black tiger shrimp, *Penaeus monodon* to improve a locally produced probiotic/bioaugmenter for controlling pathogenic *Vibrio* in Sri Lankan shrimp culture systems

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Bacillus species are the major probiotic bacteria used in both hatchery and grow-out systems of tropical shrimp culture industry to control pathogenic *Vibrio* species. Our previous works have confirmed that the use of a locally produced probiotic/bioaugmenter containing a locally isolated strain of *Bacillus subtilis* could contribute significantly in producing healthy post larvae of *Penaeus monodon* and in obtaining a profitable harvest from grow-out ponds compared to systems that did not use the product. Therefore, present study was planned to isolate different isolates of *Bacillus subtilis* to improve that probiotic or to formulate a new probiotic with improved performances. Samples of apparently healthy black tiger shrimp, *P. monodon* residing in estuaries in the North Western Province were transported to the laboratory. Gastrointestinal tract samples of the shrimp were first subjected to heat treatment (to kill non-spore forming bacteria) and isolation, purification and characterization of different isolates of *Bacillus subtilis* were carried out following standard microbiological procedures. Isolates were identified as different strains of *Bacillus subtilis* by biochemical tests in API 20E and API 50 CHB test kits (bioMerieux, France).

Out of the nine different isolates of *B. subtilis*, only three displayed high growth rate while tolerating wide range of salinity ($5gL^{-1}$ to $35gL^{-1}$) and pH (6 to 10). Antagonistic properties of those three isolates of *B. subtilis* on five species of pathogenic *Vibrio* of cultured shrimp in Sri Lanka (isolated and identified in our previous work) were investigated. The diameter of inhibitory zones produced by the new isolates of *B. subtilis* on the pathogenic *Vibrio* species ranged from 2 to 4 mm; they could be used to improve the locally produced probiotic/bioaugmentor. The selected three isolates of *B. subtilis* have been sent for the confirmation of strain type by DNA analysis.

Key words: Penaeus monodon, pathogenic Vibrio, Bacillus subtilis

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