

## Screening and evaluation of local bacteria isolated from shellfish as potential probiotics against pathogenic Vibrios

### ABSTRACT

The present study was carried out to isolate, screen and evaluate potential candidates of local bacteria isolated from tiger shrimp *Penaeus monodon* and slipper cupped oysters *Crassostrea iredalei* as probiotics in shellfish aquaculture. A total of 144 of bacteria were successfully isolated from the intestine and stomach of 20 tails of healthy adult tiger shrimp *P. monodon*, while 136 were successfully isolated from the digestive tract, gills and inner shells of 10 healthy adult *C. iredalei*. The number of potential isolates was narrowed down to two from tiger shrimp, and one from slipper cupped oyster after in vitro screening assays. The three isolates, labeled as G11, I24 and S66, were identified as *Virgibacillus* sp., *Bacillus* sp. and *Exiguobacterium* sp., respectively, using 16S rDNA gene analysis. The antagonistic ability of the isolates towards *Vibrio alginolyticus* and *Vibrio harveyi* were conducted in stagnant and liquid modes via spot lawn and broth co-culture assay, respectively. In these assays, all the potential probionts were inhibitory to both pathogenic vibrios. In the in-vivo assay, *Artemia* was used as host and treated with different concentrations of potential probionts ( $10^4$ ,  $10^6$  and  $10^8$  CFU ml<sup>-1</sup>), and challenged with *V. alginolyticus* and *V. harveyi* at  $10^5$  CFU ml<sup>-1</sup>. Respectively, *Artemia* treated with probiont G11 at all concentrations and challenged with *V. alginolyticus* had increased survival (70-80 %), which was significantly higher as compared with group with only the pathogen (20 %). Meanwhile, probiont I24 increased the survival of *Artemia* by 70 % at a concentration of  $10^8$  CFU ml<sup>-1</sup> after being challenged with *V. alginolyticus* and *Artemia* treated with  $10^6$  CFU ml<sup>-1</sup> of probiont S66 had increased survival of 90% after being challenged with *V. harveyi*. Thus, the three isolates might have potential applications as probiotics in shellfish aquaculture against vibriosis.

**Keyword:** *Crassostrea iredalei*; Potential probiotics; *Penaeus monodon*; Vibriosis