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Evidences of abundant hemocyanin variants in shrimp *Litopenaeus vannamei*

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Hemocyanin (HMC) is a multifunctional immune molecule present in mollusks and arthropods and functions as an important antigen non-specific immune protein. Our previous evidences demonstrated that *Litopenaeus vannamei* HMC might display extensive molecular diversities. In this study, bioinformatic analysis showed dozens of variant sequences of the HMC subunit with higher molecular weight from *L. vannamei* (LvHMC). Three variant fragments, named as LvHMCV1-3, which shared 85-99% nucleotide identity with that of the classical form of LvHMC (AJ250830.1), were cloned and characterized. Spatial expression profiles showed that LvHMCV1-3 had different tissue-specific distribution, which were affected by stimulation with six pathogenic bacteria, including *Escherichia coli* K12, *Vibrio parahaemolyticus*, *Vibrio alginolyticus*, *Vibrio fluvialis*, *Streptococcus pyogenes* and *Staphylococcus aureus*, with each variant fragment showing a specific stress pattern to different bacterial pathogens. Full length cDNA of LvHMCV3 was further cloned and characterized. The deduced amino acid sequence shared 92% identity with that of LvHMC, possessed a conserved structure characteristic of the HMC family and could be clustered into one branch along with other arthropod HMC in a phylogenetic tree. In addition, the recombinant protein of LvHMCV3 (rLvHMCV3) showed obvious agglutination activities against three aquaculture pathogenic bacteria including *E.coli* K12, *V. parahaemolyticus* and *S. aureus* at concentrations ranging from 31.25 to 62.5 g/mL. It also showed obvious antibacterial activity against *V. parahaemolyticus* at concentrations 0.02 to 0.5 mg/mL, and possessed the best inhibitive effects compared with those of rLvHMCV4 and rLvHMC. Co-injection of *V. parahaemolyticus* and rLvHMCV3 in *L. vannamei* showed significant decrease in mortality rate at 24-72 h after injection. Therefore, these studies suggested that *L. vannamei* had abundant HMC variants, which possessed obvious resistance to pathogenic infection and might specifically target on different pathogens in shrimp.

Biography

Yue-Ling Zhang is a PI in Guangdong Provincial Key Laboratory of Marine Biotechnology, School of Sciences, Shantou University. He received his PhD from Xiamen University, China and studied at Uppsala University, Sweden as a visiting scholar. He was a Professor and Group Leader of Department of Biology, Shantou University, China from 11/2011 to 03/2013, 05/2014 to 11/2016, and then moved to Marine Biology Institute, Shantou University, China as a Professor and Group Leader since 11/2016. He is the Director of Guangdong Province Society for Biochemistry and Molecular Biology, a Standing Director of Guangdong Province Society for Biochemistry and Molecular Biology, a Director of Guangdong Province Society for Zoology, and a Director of Guangdong Province Society for Aquaculture. His main research is focusing on shrimp immunology, especially the immune mechanisms of crustacean hemocyanin. So far, he has been the recipient of about 20 foundations as Principal Investigator. In peer-reviewed publications, his group has published 46 research papers in *J Proteome Res*, *Fish Shellfish Immunol*, *FEBS Lett*, *Dev Comp Immunol*, *Immunol Lett*, *PloS ONE*, *Mol Immunol*, *J Shellfish Res* and so on.

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