## *"Impact of Vibrio harveyi quorum sensing on the survival and the immune response of hybrid grouper larvae"*

## Ms. Shariza Azizan Assoc. Prof. Dr. Natrah Fatin Mohd Ikhsan Professor Dr. Fatimah Mohd Yusoff Institute of Bioscience Universiti Putra Malaysia

High mortality is common in grouper and white shrimp larviculture due to its undeveloped immune and, digestive systems which lead to shortages of the seed supply. The mortality is reported to be associated with disease problem such as bacterial diseases. Vibriosis caused by Vibrio sp. e.g Vibrio campbellii could affect various stages of the culture. The virulence of V. campbellii is reported to be controlled by quorum sensing (QS) mechanism through signal molecules known as Harveyi autoinducer-1 (HAI-1), Autoinducer-2 (AI-2) and Cholerae autoinducer-1 (CAI-1). Hence, this study envisioned to investigate link between QS on the virulence of V. campbellii towards grouper and white shrimp larviculture. The larvicultures were challenged with V. campbellii wild type (BB120) and maximal or minimal QS mutants (JAF483, JAF548, JMH634 and BB152). Challenged of tiger grouper larvae showed that BB120 was pathogenic to grouper larvae, causing more almost 50% mortality after 24 hours and 100% mortality after 48 hours post-challenge. Furthermore, the mortality of grouper larvae challenged with maximally active QS (QS+), JAF483 was significantly higher compared to BB120, whereas a higher survival was observed in the grouper larvae challenged with minimally active QS (QS-), JAF548. Grouper larvae challenged with autoinducer synthase triple mutant, JMH634, and Harveyi Autoinducer-1 (HAI-1) synthase mutant, BB152 also showed higher survival than BB120. Addition of HAI-1 to the BB152 showed no significance different to the larvae challenge with BB152. It is suggested to prolong the experiment to fully observe the impact. The experiment was repeated white shrimp PL20 in addition of AI-2 synthase mutant strain, MM30 and CAI-2 synthase mutant strain, JMH603 mutant strains. The results showed that the wild type V. campbellii BB120 significantly causing approximately 50% of survival rate in the control treatment after 14 days of challenge in comparison to the control without challenge (81%). Maximally active QS strain, JAF483 (QS+) and HAI-1 synthase mutant strain, BB152 also showed approximately 50% survival. Meanwhile, high survival rate (68-76%) was significantly observed in the shrimp challenged with minimally active QS strain, JAF548 (QS-), QS triple autoinducer mutant strain, JMH634, MM30 and CAI-2 synthase mutant strain, JMH603. The outcomes showed in the absence of HAI-1 autoinducer, low shrimp survival was observed. However without AI-2 and CAI-1 the shrimp showed high survival. This indicated that AI-2 and CAI-1 play a role for the virulence of V. campbellii towards shrimp postlarvae. This study provides the information that can be used to develop the probiotic as an inhibitor towards virulence of V. campbellii for aquaculture larvae.

Keywords: grouper, *Vibrio campbellii*, quorum sensing, autoinducer. *Funded by Toray Science Foundation, Japan* 

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