

Severity of white spot virus infection and WSV resistance in brood stocks of *Penaeus monodon* – identified by DNA markers

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Huge economic losses that has been occurring in global and national cultured shrimp industry due to white spot disease (WSD) could be controlled if specific pathogen free (“SPF”) and specific pathogen resistant (“SPR”) post-larvae of shrimp for white spot virus (WSV) are stocked in grow-out ponds. Present study investigated whether WSV resistant brood-stocks of *Penaeus monodon* are available in the coastal sea of Sri Lanka using 317 bp and 71 bp microsatellite DNA markers for the production of “SPF” and “SPR” shrimp post-larvae.

Brood stock samples of black tiger shrimp, *Penaeus monodon* were collected from eight major brood stock collecting sites of the coastal sea of Sri Lanka. DNA from a small piece of a pleopod from each shrimp was extracted and the severity of WSV infection was examined by PCR using IQ200 TM WSV detection kit; extracted DNA was employed to identify WSD-susceptibility and WSD-resistance using 317 bp and 71 bp microsatellite DNA markers. New forward and reverse primers were designed according to 71 bp microsatellite DNA sequence obtained from the data base of National Centre of Bio Informatics for *P. monodon*.

Reaction with IQ200 TM WSV detection kit generated three DNA bands at 720 bp, 310 bp and 210 bp from the WSV genome based on the severity of infection. Brood shrimp collected from Marawila, Chilaw, and Hendala sea recorded a very severe WSV infection that produced all three DNA bands with 10^5 viral particles. Moderate severity with DNA bands at 310 bp and 210 bp were observed in samples obtained from Negombo sea having 10^3 to 10^5 WSV viral particles. Brood shrimp collected from Beruwala, Pottuvill, Mullattivu and Valahchenai recorded a low severity of WSV infection producing DNA band only at 210 bp with 10 to 200 viral particles. Mean percentage prevalence of WSV resistance recorded for brood shrimp collected from Beruwala sea was significantly higher (87.5%; $P < 0.05$) being positive for 317 bp DNA band ; those shrimp had significantly low WSD-susceptibility (12.5% mean prevalence being positive for 71 bp DNA band). Highest mean percentage prevalence of WSV-susceptibility was recorded for brood shrimp collected from Chilaw sea (78.1%) while mean prevalence of WSV resistance of the same samples was the lowest (21%). WSV resistant brood shrimp were found in the Sri Lankan coastal sea that could be used to produce “SPF” and “SPR” post larvae which could be a breakthrough in cultured shrimp industry of Sri Lanka. The prevalence of disease resistance in brood stock samples collected from eight major brood stock collecting sites according to both 71 bp and 317 bp microsatellite markers, when ranked in descending order was Beruwala, Pottuvill, Valahchenai, Mullattivu, Marawila, Negombo, Handala and Chilaw. Accession number KX 156833 was received for the genomic sequence of microsatellite DNA marker designed for screening WSV resistance of *P. monodon* population living in Sri Lankan coastal sea.

Keywords: WSV resistance, *P. monodon* brood shrimp