

Ekachai Duangjai 2015: Effects of Salinity on Hormonal Changes of Previtellogenesis and Sperm Quality in The Pacific White Shrimp (*Litopenaeus vannamei*). Doctor of Philosophy (Aquaculture), Major Field: Aquaculture, Department of Aquaculture. Thesis Advisor: Assistant Professor Oraporn Meunpol, Ph.D. 184 pages.

Female *L. vannamei*, (40 g.bw, 120 days approx.) with previtellogenic ovaries were reared in cages (5x5 m<sup>2</sup>) installed in two earthen ponds; one filled with 15 ppt and others filled with 30 ppt. Haemolymph, hepatopancreas and ovaries of shrimp were collected monthly for hormone analysis. P<sub>4</sub>, E<sub>2</sub> and PGF<sub>2α</sub> levels in shrimp haemolymph were measured by either RP-HPLC or RIA. Biological marker (primer) specific for 314 bp of FAMeT and 357 bp of Vg gene modified from NCBI has been developed from hepatopancrease of female *L. vannamei*. Vg and FAMeT gene expression level using 1<sup>st</sup> cDNA synthesis from hepatopancreas and ovary tissues were determined by RT-PCR. The results showed that the levels of Vg and FAMeT gene expression in ovaries and hepatopancreas of shrimp reared in 30 ppt were significantly higher than shrimp reared in 15 ppt. The mean expression level of Vg and FAMeT gene in hepatopancreas of shrimp was higher than in ovaries. Similarly, changes of P<sub>4</sub>, E<sub>2</sub> and PGF<sub>2α</sub> levels and also GSI of shrimp reared in 30 ppt were significantly higher than shrimp reared in 15 ppt ( $P < 0.05$ ). In conclusion, it was clearly shown that salinity played an important role over shrimp ovarian development which could be proved at different levels i.e. morphological changes (GSI), hormonal changes and gene expression levels changes. This study was also the first to use comet assay for gamete quality determination of males shrimp by compared the traditional techniques and comet assay technique to assess sperm quality obtained the different numbers of nuplius of four groups reared two months under 30 ppt conditions. These techniques also use to study the sperm quality obtained from different sea water (15 and 30 ppt) and rearing condition (cement and earthed pound). The results demonstrated that average total sperm counts, sperm dead (%), sperm diameter (%) and spermatophore formation measured by traditional techniques showed no significantly different results ( $P > 0.05$ ). Whereas comet assay as Healthy cell, Comet Length, Tail Length, %Tail Moment, %DNA in Tail and Olive Moment displayed significantly different results ( $P < 0.05$ ). The results showed that the biological marker (P<sub>4</sub>, E<sub>2</sub> and PGF<sub>2α</sub> levels and also Vg and FAMeT gene expression level) can use to determine the change of gonad development of females *L. vannamei*. The results also showed that comet assay is more powerful than traditional techniques due to its ability in measuring at deeper level i.e., DNA quality of sperms while the latter techniques can reveal only superficial aspects of sperm quality.

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Thesis Advisor's signature