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Impact of White spot syndrome virus disease on gills in Shrimp, *Penaeus monodon*

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ABSTRACT

Protozoan parasites cause problems in Penaeid shrimp culture and major impact on the shrimp farming industry.White spot syndrome virus (WSSV) is a major shrimp pathogen that also infects *Penaeus monodon* species was selected. Histological changes in the gill cells were studied. Histological techniques using paraffin embeded tissues as well as frozen tissues were used for identification of WSSV infection. Histological manifestation such as gill cells could be detected. The gill cells were marked and characteristic of WSSV infections were observed. The present investigation was focused on the virus infecting shrimp. The microscopic examinations of the histological preparation are presently used to detect WSSV zoothamnium.

Figures : 05 References : 27 Table : 01

KEY WORDS : Gills, *Penaeus monodon,* Shrimps, WSSV.

Introduction

Diseases seriously threat the shrimp farming. Disease is one of the limiting factors for the aquaculture industry throughout the world. Asia region is the center of fishing and aquaculture activities with top fish producing countries like Japan, China, Thailand, Philippines, Malaysia and Vietnam[8, 9]. India is the second largest aquaculture producer in the world with production in 1994 of 1.6 metric tons[7]. Disease is one of the limiting factors for the aquaculture industry throughout the world. The diseases among Penaeid are somewhat dependent upon the system of culture followed. In the case of high density culture as intensive and semi-intensive, ponds encourage the development and transmission of many diseases[6, 13].

The zoothamnium infection in the gills of *Penaeus setifeus* and the brown shrimp *P. aztecus* throughout Mississippi coastal and adjacent coastal water, the infection was triggered by the water quality[16]. The infection of wild crustaceans is due to the release of untreated liquid and solid waste from shrimp importing and processing plant directly into the coastal water[2]. It confirmed that higher shrimp densities lead to the spread of pathogens and in the case of polyculture the disease spread is limited[12]. White Spot Syndrome was detected from the crab *Parase sarmapictum*. It is one of the most common residents of shrimp farms and it might transmit virus to shrimp by acting as a vector.The *P. monodon* of WSSV in both hemolymph and tissues corresponded to the severity of infection determined by histological evaluation. The histological studies demonstrated that a viral agent infected the epidermis of the diseased shrimp and the tissue degradation characterized by hypertrophiednuclei with inclusion[22, 26]. WSSV was particularly prevalent in gills, followed in order of decreasing prevalence by hemolymph, stomach, pleopods, heart, integument, periopods, eyestalks and hepatopancreas[14]. The present study was impact of white spot syndrome virus disease on gills of shrimp *P. monodon*.

Materials and Methods

Infected White Spot Syndrome disease shrimp *Penaeus monodon* was used in the present investigation. The samples were collected from various aquacultures in Sindhudurg district in Maharashtra, India. The *P. monodon* shrimps were from aquaculture farm and hatcheries through different semi Government as well as registered private sectors in Sindhudurg district. Collected shrimps were properly preserved on the spot. The specimen animals were brought in the laboratory. The shrimps were then further processed for study. The infected portion also dissected and infected portion of gills tissues were fixed

TABLE-1:Detection of the infected gills organsWhite Spot Syndrome Virusof the shrimp, *P. monodon*.

Name of the shrimp species	Natures of culture	Organs of *P. monodon* Shrimps		Investigation for tissue
Penaeus monodon	Aquaculture(Pond)	Gills	lamellae	Positive
			Filaments	Positive
			Rackers	Positive

with Davidson's fixative for 24 to 48 hrs. Then they were transferred and preserved in 50% ethyl alcohol for subsequent histological prepration and analysis. These samples were processed and sectioned following paraffin embedding using the standard methods[2]. The histological techniques were used.

Fig.1: WSSV infected on gills of *P. monodon*

Fig.2: WSSV infected gills in the *P. monodon*

The tissue of the gill *P. monodon* was sectioned at 4-5 µm in thickness. Sections were stained with haematoxylin and eosin. The slides were further dehydrated in ascending grades of alcohol and finally given two changes of absolute alcohol. Slides were then cleared in xylene and mounted[17] DPX. Subsequent gross significance was examined by light microscopy for histological typical studies of White Spot Syndrome Virus (WSSV) infection was evaluated.

Results and Discussion

In the present study infected shrimp *Penaeus monodon* was observed and selected (Fig.1). The *P. monodon* shrimps of the body part gills were observed through light microscopic analysis (Figs. 2,3,4 and 5). The body parts of the *P.monodon* are shown positive for White Spot Syndrome Virus (WSSV) (Table-1).*P. monodon* shrimp during the monsoon period WSSV infection was recorded.The *P. monodon* shrimps signs and symptoms were observed. The WSSV

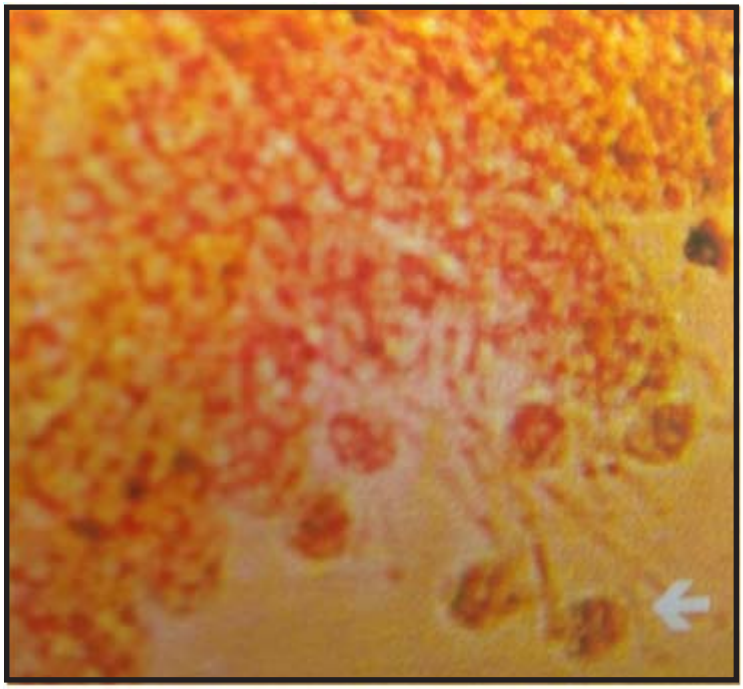

Fig.3: Ectoparasites infection in the gills

disease spread very rapidly and affected almost all the shrimp in the culture. The infected shrimp died within 5 to 15 days, spots were very clear in the gills of the infected shrimps.

This has been confirmed in the report that WSSV infection was environment factor like rain. In the case *P. indicus*, *Metapenaeus* species important, species being cultured by Indian shrimp farmer but in *P. monodon* the lowest infection rate observed was 14% in male and 25% female for being cultured world wise[1, 3, 5.21,24]. WSSV was detected in *P.joponicus* in the culture facilities, wild and experimentally infected shrimp. The tiger shrimp *P. monodon* and *P.indicus* from three systems (culture, wild and experiments) showed infection (microscopic white spot) [17, 25]. It was detected in culture facilities, wild and auto entry in the *P. monodon* culture ponds[27]. On the other infected *P.monodon* was also found in the crab fattening *Scylla tranquebarica* pond and in *P. indicus* culture ponds. The Macrobrachium rosenbergiii of tolerance were WSSV infection[14, 20, 23,26]. During heavy rains the extensive outbreaks of WSSV in grow out ponds was recorded. The other environmental parameters like pH, Temperature and Salinity *etc.* however, do not significantly influence the rate of infection[11].

In the present study brooder of *P.monodon* were selected for monitoring.The infection of the wild *P. monodon* brooders in Southern Taiwan was reported that the percentage of infection (PCR confirmation) among male was 40% and female 12.5%. In the present study the females were found more infected than the males, the highest percentage of infection recorded in the study being 14% in male and 25% in female. Both in the brood stock bank and hatchery (maturation phase) the shrimps were fed with crab, bivalve, polychaete worm and shrimps for the growth and fast maturation of brooder [10,14,19,21]. During this period, there is a chance of WSSV infection through shrimp and crabs. Recent study showed that WSSV infects almost all the crustaceans both in captivity and in the wild. Polychaetes are the best brood stock feed. WSSV infection through food could be avoided through the exclusive use of non-crustaceans[2].

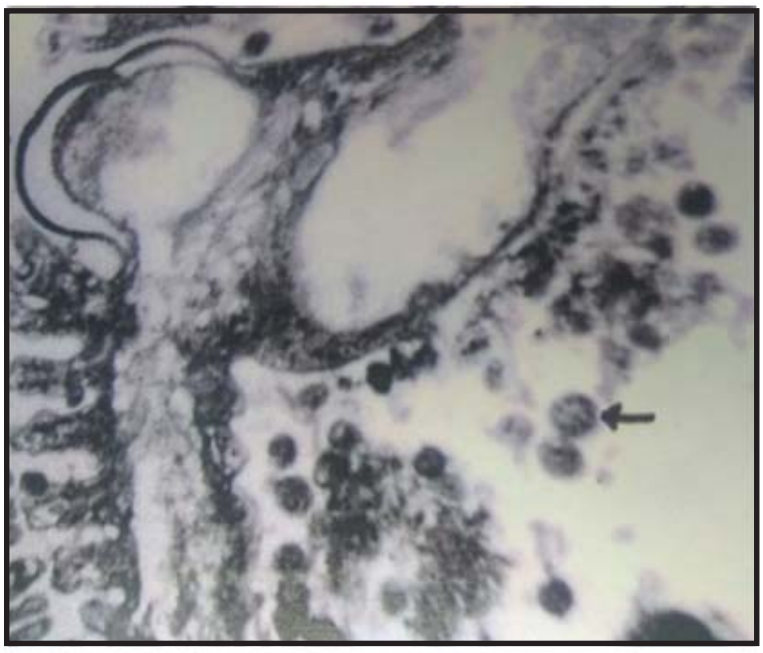

Fig.4: Gill region infected of *P. monodon*

Histological studies of the present study showed that the gill cell and abdominal muscles cell characterized by hypertrophied nuclei and eosinophilic to basophilic inclusions and cellular necrosis[13]. Protozoan fouling in the gills of the infected shrimps was reported. In the present study protozoan fouling was detected in almost all the viral infected shrimps. Algal fouling was also observed in the gills of *P.monodon*[4, 18].

If water quality is not maintained properly, shrimps become susceptible to disease. Shrimps respond to changes in every water quality parameters. Temperature plays an important role in the metabolism of shrimps. The optimum temperature for grow-out pond was 25 to 30°c, but temperature increased 33 to 37°c to the summer season [3]. *P. monodon* is a euryhaline animal, which can tolerate wide range of salinity. The salinity plays a major role in the growth of shrimps. The optimum salinity in all the ponds ranged between 24 to 35.7 ppt and salinity was also low 15 to 22ppt. the pH

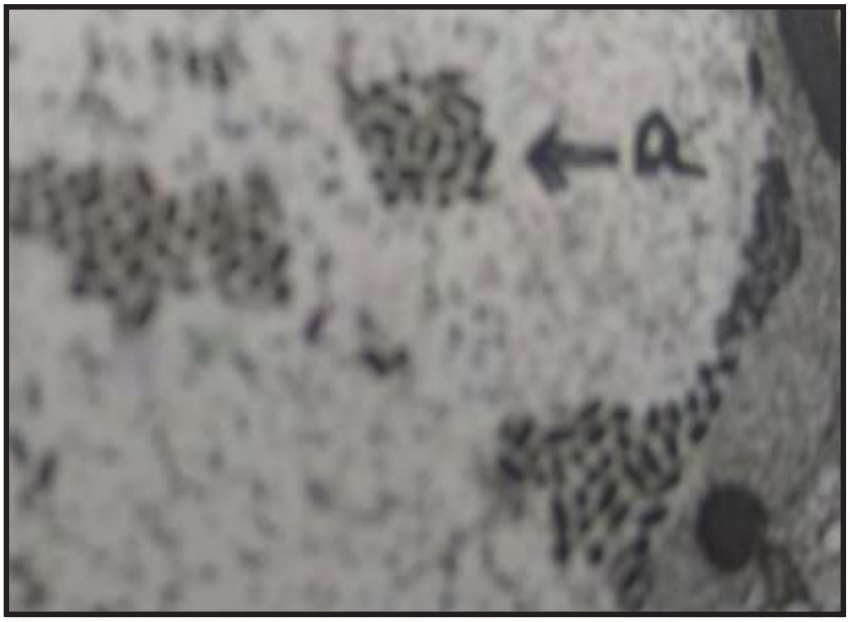

Fig.5 : WSSV disease infected in the gills lamellae of *P. monodon.*

fluctuation is not a major threat to the culture animals because the buffering capacity of the brackish water maintained the pH equilibrium. Low pH increases the toxicity of nitrite to culture organism and higher pH leads to increase in unionized ammonia, another toxic material to the shrimps. The dissolved oxygen level less than 3ppm is detrimental to the shrimps. Low dissolved oxygen content increases the ammonia concentration[15]. The water transparency was monitored by secchi disc and it was optimum level of transparency from 25 to 45 cm. The nutrients, nitrate and phosphate are the limiting factors for the phytoplankton production[3, 12]. It is concluded that the impact of WSSV infection affects the *P.monodon* internal tissues and different part of organs appearance gill lamellae, gill racker, gill filaments and various segments muscles are affected.The aquaculture industries which determine the success of shrimp *P. monodon* culture and its efficient management are essential for aqua culturing farms and hatcheries.

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