



CHITINOLYTIC BACTERIAL DISEASE INFECTED IN *SCYLLA SERRATA* (FORSSKAL 1775) FATTENING FARM OF TAMIL NADU

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ABSTRACT

Chitinolytic bacterial disease is responsible for great economic losses in the commercial mud crab fattening in the farm of Tamil Nadu. In the present study, water quality parameters analysed for normal and infected farm of fattening period. The normal farm observed in the minimum temperature 3rd days and maximum was 45th days. The infected farm also observed in the minimum temperature of 27th days and maximum 39th days. The water temperature appears to have a strong influence on the observed temporal patterns of the recent shell disease outbreak. The histological examinations of normal and diseased mud crab *S. serrata* gill lamellae were examined. The chitinolytic bacterial disease affected in the *S.serrata* ventral surface showed. The infected gill lamellae showed degradation of the epithelial layer and hemocytic nodules were observed the hemal sinuses

Keywords: *Scylla serrata*, water quality parameters, chitinolytic bacteria disease and Histology

INTRODUCTION

Mud crab, *Scylla serrata* (Forsskal, 1775) is a large portunid crab found in greater abundance in all estuaries, coastal lagoons and near shore waters of India. The mud crabs belongs to the genus *Scylla* and family Portunidae is one of the exotic aquaculture species and economically important crustacean species. Mud crabs has been practiced for several decades in many Asian countries including India (Kathirvel et al., 2004). In India, mud crab farming mainly depends on fattening of soft shelled or water crabs. This is probably the simplest form of aquaculture practice where recently molted crabs are held in confinement for a short period to enhance marketable attributes. The mud crab fattening has been developed rapidly in past few years in worldwide and increase the production. However, the mud crab fattening outbreaks of shell disease were facing economic losses in Pemalang district, Central Java, Indonesia and India (Lavilla-Pitogo, & de la Peña, 2004; Austin, B., & Allen-Austin, 1985).

Shell disease syndrome (SDS) is one of the major problems affecting freshwater and marine crustaceans species (Sindermann 1989). It is a bacterial disease such as filamentous bacterial diseases, luminescent bacterial diseases and shell diseases or chitinolytic bacterial disease has been reported in mud crabs (Lavilla-Pitogo, de la Pena 2004). Chitinolytic bacterial disease is a common disease of crustaceans species that cause various types of erosive lesions on the shell (Johnson, 1983; Sindermann and Lightner, 1988). Chitinolytic bacteria is a gram negative bacteria such as *Vibrio* spp., *Aeromonas* spp., *Pseudomonas* spp., *Flavobacterium*, *Moraxella*, *Pasteurella* and *photobacterium* are involved in the infection (Wang 2011). The shell degradation of crustacean species in the infection of chitin outer layer the exoskeletal (cuticle) can be accompanied by melanisation of the affected region (Vogan et al., 1999). The numerous shell disease reports on the blue crabs and other crustaceans (Rosen 1967) with comprehensive description of the shell lesion pathology. The shell lesion cell affected crabs were histological studies of gills, hepatopancreas and heart (Vogan et al., 2001). The diseases not believed to be fatal in initial stages, which the mortality crabs were incomplete moulting and shell lesion infected area entire to the pathogenic microorganism (Baross & Tester, 1978; Vogan et al., 2001 & Smolowitz et al., 1992). The disease has been reported in many crustaceans species of economic importance (Sindermann, 1989) and in association with stressful environments, such as intensive aquaculture, impounded population and polluted natural environments (Sindermann, 1990; Taylor, 1948; Gopalan and Young, 1975; Young and Pearce, 1975; Noga 1991). In this study was no report of chitinolytic bacterial disease in the *S. serrata* fattening farm of Tamil Nadu.

MATERIALS AND METHODS

Study area:

M.G.R thittu (11° 28' 11.4" N; 79° 46' 46.9" E) is a coastal village between Cuddalore district, Tamil Nadu, India. In this area, a total of 15 crab fattening pens farms extensively cultured *S. serrata* between 2015.

Sample collection:

The shell disease affected crabs (*S. serrata*) of 300-400 g of body weight which were collected from the pens fattening farms during December 2015. For each sampling, 18 infected crabs were collected and brought to the laboratory for further investigation.

Water quality parameters of pens fattening farm water:

The water quality parameters viz., temperature, salinity, pH and DO of pens fattening farms were analysed according to standard methods (APHA, 1998).

Histopathological study:

The normal and shell disease infected gill tissue samples were preserved in the Davidsons fixative was kept at room temperature 48 h after transferred to 70% ethanol. The organs were dissected and arranged in embedding cassette. The tissues were processed with different grades of ethanol and other reagents as described by Bell and Lightenr (1998). The paraffin embedded tissues were cut into section of 5 μm thickness using a rotary microtome and were stained using hematoxylin and eosin (H&E). Finally the sections were mounted with DistyrenePlasticzer Xylene (DPX). The slides were observed under a light microscope (Magniis Microscope).

RESULTS

Gross morphologic finding:

The severe lesion on the *Scylla serrata* infection was shell disease or chitinolytic bacterial infection on abdomen male crab of the ventral side (crab weight 620 gm; carapace width 15.4 cm). (Fig.1).

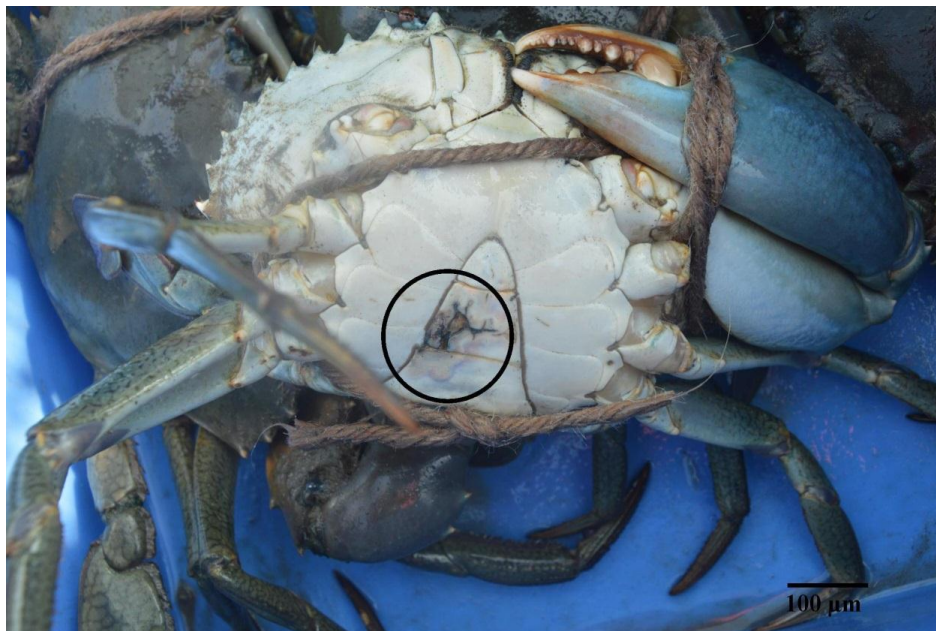


Figure 1: Chitinolytic bacterial infection on abdomen of male crab (circle).

Histopathology:

The histological investigations of normal and infected gill lamellae were observed in the *S. serrata*. The central axis bears densely arranged numerous paired leaflets, each of which represents a cavity filled with hemocyte and enveloped in a thin cuticle. The cuticle, each leaflet is lined with a layer of the simple respiratory epithelium. Inside the each leaflet, at certain intervals, there are columnar, strongly extended, pilaster cells, which join to other center of the leaflet lumen to make up isolated chambers (Fig. 2A). The severe lesions of gill lamellae infected with hemocyte nodules were observed occluding the hemal sinuses and degradation of the epithelial layer (Fig. 2B and C).

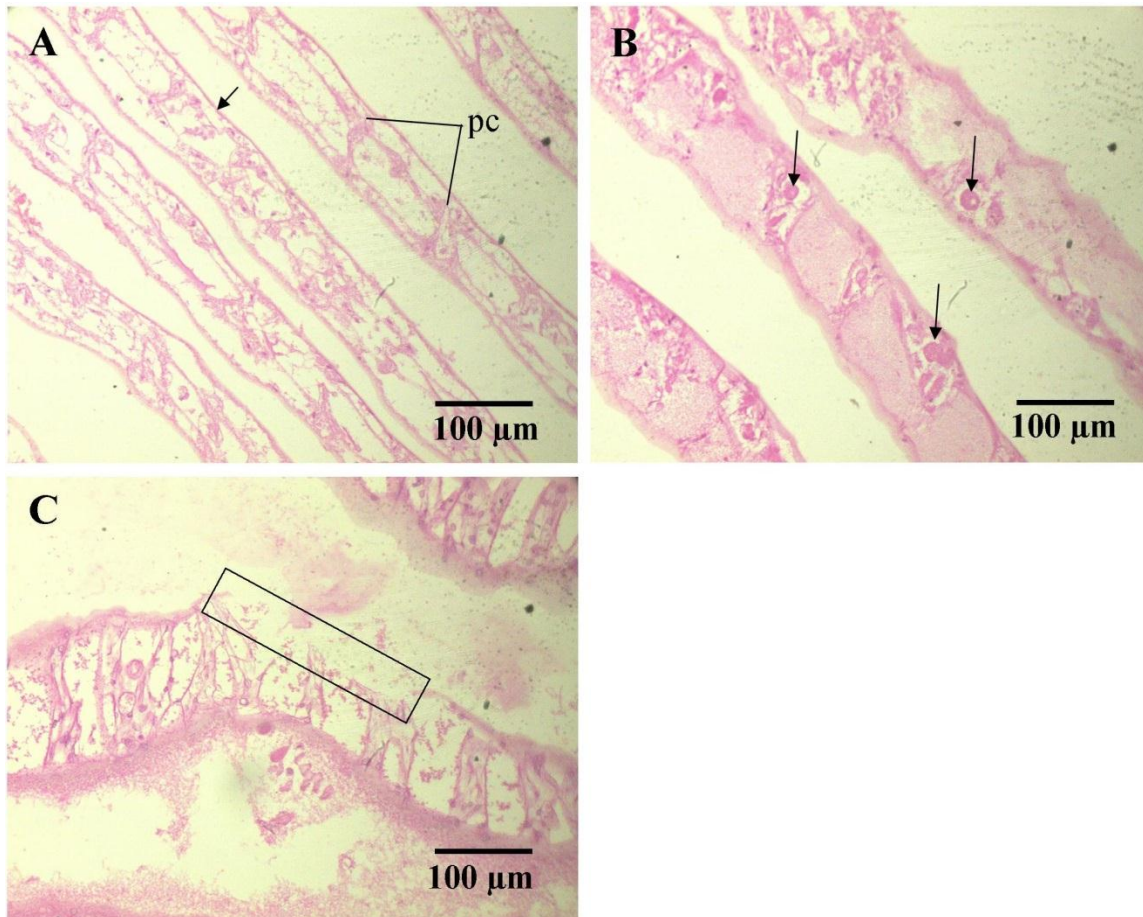


Figure 2: Histology section of normal *S. serrata* gill lamellae present in the epithelial layer (arrow), pilaster cells (pc). B) Shell disease infected gill lamellae in hemocytic nodules (arrow) and C) degradation of epithelial layer (frame).

Water quality parameters:

The water quality parameter such as temperature, salinity, pH and dissolved oxygen were monitored since 3rd days of culture. The temperature in the normal pond ranged between 27.2 to 29.8°C. The minimum temperature was observed on 3rd days of culture and the maximum was recorded 45 days of culture. In the

shell disease infected pens fattening farm also observed in the minimum temperature was observed on 27 days of culture and the maximum was recorded 39 days of culture. (Fig. 3).

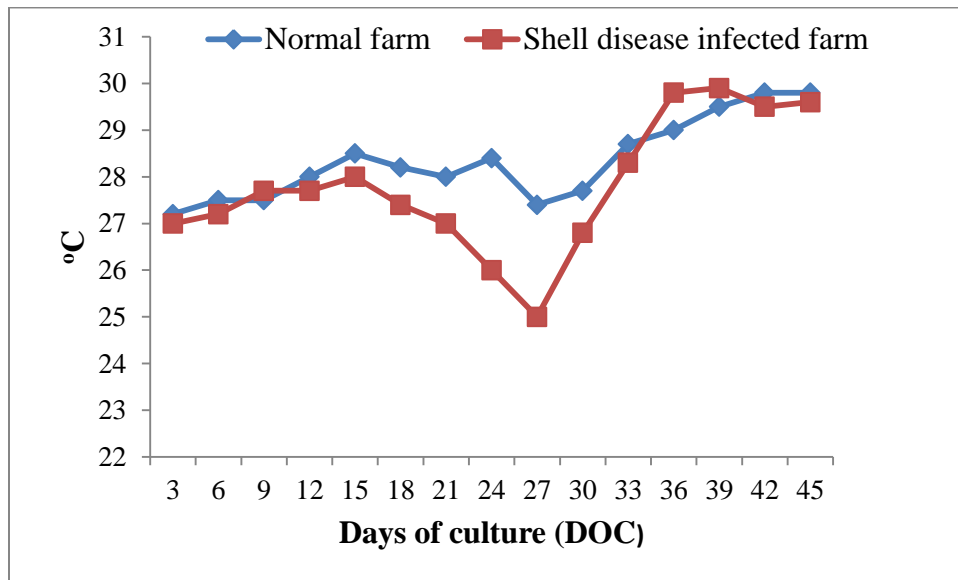


Figure 3: Temperature variation in the *S. serrata* pens fattening farm

The pH value in the normal pond varied from 7.6 to 8. The minimum pH value was recorded on 18th days of culture and maximum was observed on 36 days of culture. In the shell disease infected pens fattening farm the pH value varied from 7.5 to 7.9. The minimum pH value was recorded on 9th days of culture and the maximum was recorded on 3rd days of culture (Fig. 4).

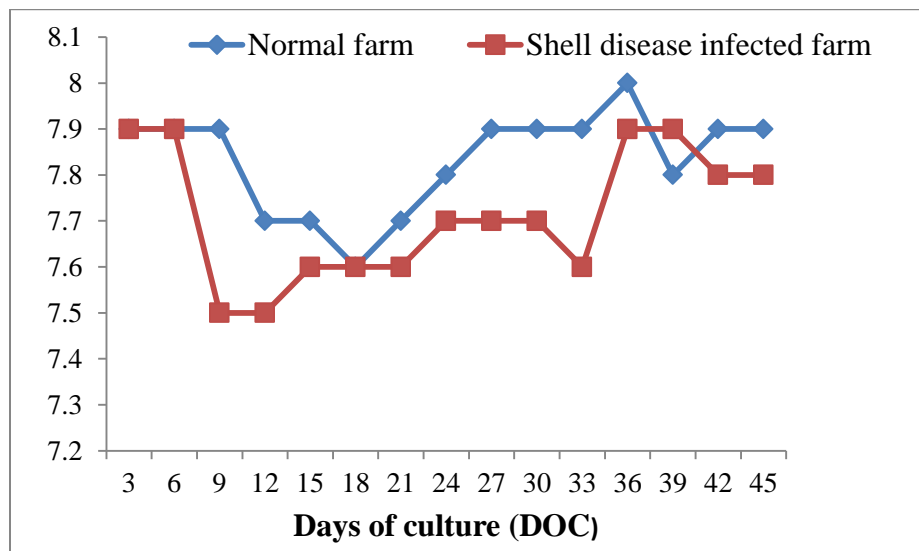


Figure 4: pH variation in the *S. serrata* pens fattening farm

The salinity in the normal pond ranged between 20 and 28.5 ppt, the minimum salinity was noted on 3rd of culture and the maximum was recorded on 45 days of culture and in the shell disease infected pens fattening farm similar trend was noted and slight variation found during culture period (Fig. 5).

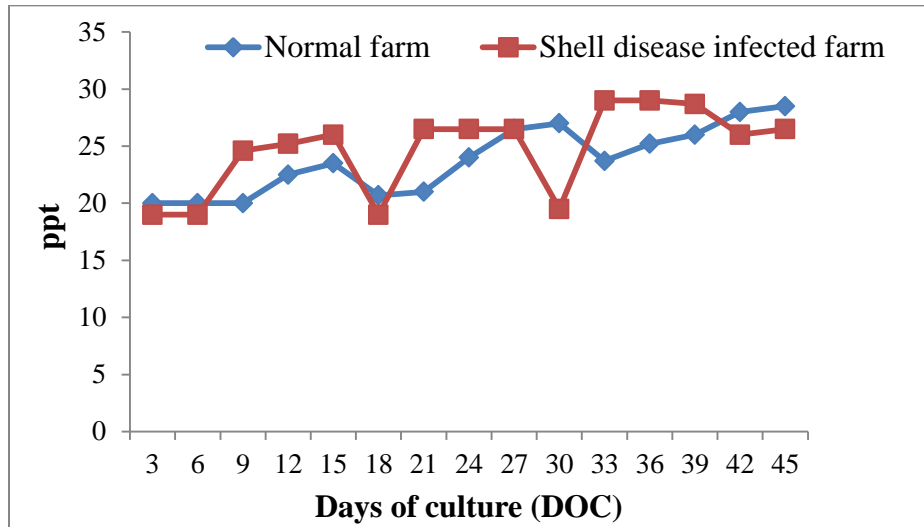


Figure 5: Salinity variation in the *S. serrata* pens fattening farm

The dissolved oxygen concentration level in the normal pond was ranged between 4.5 and 4.8 mg/L. The minimum dissolved oxygen concentration was recorded on 36 days of culture and the maximum was observed on 45 days of culture but in the shell disease infected pens fattening farm the dissolved oxygen concentration level was varied from 4.2 to 4.8 mg/L. The minimum concentration was recorded on the 30 days and the maximum was on 39 days of culture (Fig.6).

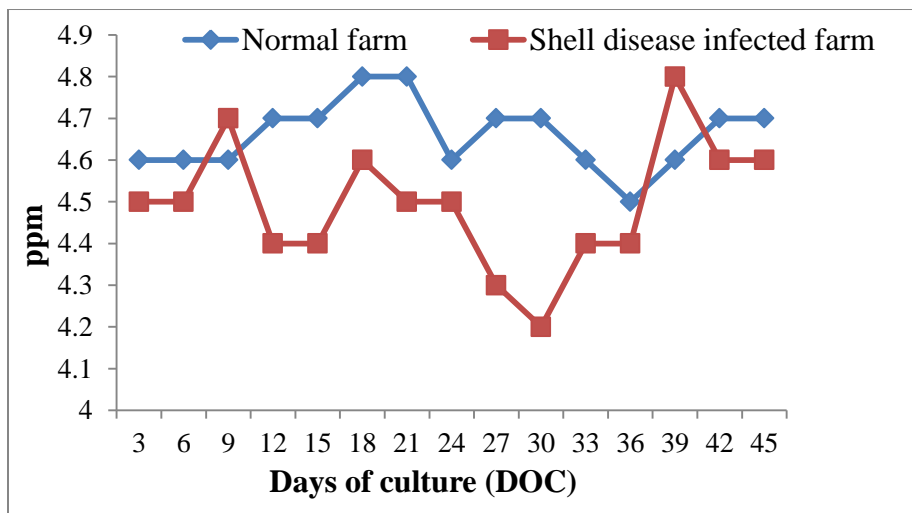


Figure 6: DO variation in the *S. serrata* pens fattening farm

DISCUSSION

The previous study of 100% survival was recorded in the mud crab *S. serrata* without acclimation at average 32.6 ppt, which was indicating the euryhaline nature of the crabs (Nair et al., 1974; Marichamy 1996). The mud crabs are highly tolerant to varying salinity conditions ranging between temperature 26 C to 32°C, pH 8.0 to 8.5, salinity 10 ppt to 34 ppt and dissolved oxygen content should be more than 3 ppm (SEAFDEC, 1997), so brackish water would be ideal for mud crab grow out culture and fattening system. The mud crab species *S. serrate* is known to occur in water bodies having a range of salinity from to 8 ppt to 45 ppt reported by (Jones and Sujansinghani, 1950). Several attempts at commercial culture of mud crab have been made but low survival has been the major constraint to commercial operation (Gillespie and Mann, 1991).

Chitinolytic bacterial shell disease is a serious disease problem of mud crab *S. serrata* fattening system in Tamil Nadu. In this study two different pens were study from brackish water. The pens preparation, stocking densities feeding were similar to the both pens farm. The water quality parameters of the normal and chitinolytic bacterial disease infected pens farm were similar but variation was occur in temperature. In case temperature was decreased in the growth of bacteria suspected to cause of shell disease. Temperature has a strong influence on the growth and reproductive cycles of lobsters (Waddy et al., 1995), thus impacting the length of intermolt duration and subsequent exposure to disease-causing agents. In addition to affecting regional patterns of shell disease prevalence, water temperature appears to have a strong influence on the observed temporal patterns of the recent outbreak. We propose that our data illustrate a lag in the relationship between water temperature and disease incidence, due to the correspondence of warm winter water temperatures and the development over time of disease symptoms. The visible symptoms of shell disease, pitting and erosion of the dorsal and ventral side.

The previous study was shell disease severity in the *Cancer pagurus*, severe lesion of infected dorsal carapace in the polluted environment (Rosen, 1967; Comely & Ansell, 1989; Smolowitz et al., 1992). The surveys of shell disease in observing studies have relied entirely upon gross evaluation (Sandifer& Eldridge, 1974; Sindermann, 1989). Our studies suggest that infected mud crab diagnosis of gross observation might be used as a crude indicator of shell disease infected in the ventralsurface of the severe lesion and blockcolour. Further, study of histological investigation in the internal organs.

The severe shell disease revealed histopathological changes of several internal organs and tissue has been reported (Vogan et al., 2001; Smolowitz et al., 1992; Ayres & Edwards 1982; Ryazanova, 2005). The increased nephrocytes number and dimension in *S. serrata*, along with necrotic and degenerative changes in the gills and hepatopancreas, respectively, have been frequently observed as a consequence of polluted environment exposure (Stentiford et al., 2003). The histological changes in the internal organs of gill cells in the red king crab infected for shell disease in crustaceans (Brock and Lightner 1990). The present study was adhesion of hemocyte nodule formation and destruction of the epithelial layer in the gill lamellae of *S. serrata*. Theshell disease might be directly due to systemic bacterial infection of the infected ventral surface. To our

knowledge about the marine environmental conditions that the lead to disease and shell lesion infected area entire to the pathogenic microorganism are capable of degradation of the chitin.

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