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Occurrence of black gill disease in relation to stocking density and some major prevailing hydrological parameters of *Penaeus monodon* in Northern Odisha, India

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ABSTRACT

Occurrence of black gill disease in Penaeus monodon was studied during 2009 - 2010 in the coastal grow-out ponds of northern Odisha. The disease was characterized by black coloration and destruction of gills due to which lamellae became atrophic and collapsed. In severe cases, the disease led to death of shrimps affecting the yield. The stocking density, area of the ponds and hydrological parameters were studied simultaneously with shrimp sampling. Results showed that salinity of the water was positively and significantly correlated with length and weight of the shrimp (r = 0.213, 0.235 at $P \le 0.05$) and stocking density was significantly correlated (r = 0.807, & r = 0.836, $P \le$ 0.01) with area of the ponds and affected % in both the years. Survival % showed positive correlation (r = 0.485, $P \le$ ≤ 0.01) with area of the ponds. Hence it was concluded that stocking density directly affected the disease affected percent.

Key words: Black gill, shrimp, stocking density, hydrological parameters

INTRODUCTION

Coastal aquaculture has been identified by the Government of India as high potential area for increasing fish and shellfish production that achieves economic and social benefits. India is with over 8100 km of coastline, vast stretches of estuaries or backwaters, lagoons which provide enormous opportunities for brackish water shrimp farming. During early nineties due to proven technology black and white tiger shrimp (*Penaeus monodon* and *Penaeus indicus*) cultured with large scale growth of shrimp farms and hatcheries which witnessed in a short span of five years [1].

In Odisha, the shrimp culture is being practiced in the coastal ponds for several decades, especially in northern parts of the state. *Penaeus monodon* is an economically important shrimp, cultured in the coastal ponds of northern Odisha along Bay of Bengal in the districts of Balasore and Bhadrak. Intensification of shrimp farms and unscientific management strategies overwhelmed the shrimp industry in the state. However mass mortality due to microbial infection is a bottleneck affecting the production. Most commonly found infection is black gill disease caused by several factors resulting in death of infected samples due to destruction of gills. Further the destruction causes blockage of gas exchange through gill lamellae leading to suffocation [2]. The black gill disease is characterized by the presence of black spot and necrosis in the gills and later get collapse at the onset of the disease [3]. The black spots are caused by the inflammatory action resulting activation of prophenoloxydase which converts phenol to melanin [4]. At the initial stages of development of the disease the gills are known to discoloration and

gradually lead to black color [5, 6 and 7]. In the year 2009 and 2010, black gill disease was frequently reported in the coastal ponds of Northern Odisha at the grow-out stage affecting shrimp production. Not much attention had been given to this disease, and its occurrence, cause and effect in yields of shrimps in the coastal ponds of Northern Odisha, however some scattered work was found in other regions of India. Hence the present work was designed to study the area of the farmed ponds, their stocking density and black gill disease affected percent of *Penaeus monodon* along with some hydrological parameters.

MATERIALS AND METHODS

Samples of *Penaeus monodon* were collected from ten ponds from S. R. Aquafarm Pvt. Ltd. present in Balasore district (site-1) and Albatross Aquafarm Pvt. Ltd. present in Bhadrak district (site-2) fortnightly from April 2009 to July 2009 and April 2010 to July 2010 for examination of the shrimp condition. Black gill affected specimen were identified and their number was recorded along with stocking density and area of the ponds. Gills from the shrimps with and without infection were stored at -20° C for morphological analysis. Hydrological parameters such as salinity (by salinometer TDS-10 pen type), water temperature (by mercury centigrade thermometer, 0.1gradation), pH (by digital pH meter), transparency (by secchidisc, cm) dissolve oxygen (DO) (Wrinkle's volumetric method), total alkalinity (by methyl orange indicator) and hardness (Ca) (EDTA method) was recorded simultaneously. The length and weight of the samples were also recorded. Sampling was done to analyse growth and disease conditions of shrimp fortnightly using cast net.

Data analysis

The data were analysed by one way Analysis of Variance (ANOVA) at 1% and 5% level of significance by using SPSS (statistical package for social sciences) version 16.0.

RESULTS

During the study period *P. monodon* samples with black gill affected condition were collected in live state from intensive farms of the study site. The external clinical sign of naturally infected specimen was black colour of the gills. The gills were found to be completely black in color (Fig.1) and the lamellae were observed to be necrotic, atrophic and collapsed. The gill lamellae of the affected samples were studied morphologically by mounting the lamellae and the slides showed the presence of fungus Fusarium species. The infected shrimps were found to be weak, lethargic and had difficulty in respiration, thus found at edges of the ponds in shallow water. In severe cases the shrimps were deposited with silt on carapace and body surfaces and exhibited heavy fouling. Some were observed with erosion in appendages. The area of ponds in Balasore (site-1) and Bhadrak (site-2) are represented in fig.3 and 4, which were ranged between 3000 to 5500 square meter. The relative stocking density was found to be 55000, 55000, 65000 and 60000 for 5 ponds respectively in 2009 and 55000,55000,65000,65000 and 60000 for the same ponds in 2010 of site-1. The water temperature varied between 28.94 $\pm 2.16^{\circ}$ C in Pond-3 of Balasore (Site-1)(2010) to 31.28 $\pm 2.23^{\circ}$ C in Pond-4 of district Bhadrak (Site-2)(2010). The average temperature of both the years was found to be 29.15 $\pm 2.11^{\circ}$ C (Fig.2). Analysis of variance did not show any significant difference of temperature between the ponds (F =0.038, P ≤ 0.01).



Fig.1 Black gilled P.monodon (Site 2)

Fig.2 Black gilled P.monodon (Site 2)



Fig.3 Area of ponds in shrimp cultured ponds in site 1 (2009-2010)



Fig.4 Area of ponds in shrimp cultured ponds in site 2 (2009-2010)

	Temp	DO	pН	Salinity	Transparency	Alkalinity	Hardness	Ca	length	weight
Temp	1									
DO	481**	1								
pН	788**	.464**	1							
Salinity	014	.177	068	1						
Transparency	.690**	457**	654**	.149	1					
Alkalinity	749**	.528**	.655**	.157	581**	1				
Hardness	.623**	047	608**	.189	.504**	449**	1			
Ca	.652**	202*	650**	.028	.476**	580**	.679**	1		
length	.012	.079	023	.213*	.026	.024	.042	.060	1	
weight	006	.093	013	.235*	.020	.055	.040	.045	.995**	1

Table 1 Correlation coefficient of hydrological parameters with length and weight of samples

**Correlation is significant at the 0.01 level (2-tailed). * Correlation is significant at the 0.05 level (2-tailed).

The dissolved Oxygen content varied between $3.98 \pm 0.38 \text{ mg} \Gamma^1$ in Pond-2 in Site -2 to $4.17 \pm 0.39 \text{ mg} \Gamma^1$ in Pond-1 and 8 in 2009 (Site-1). The average value of DO was found to be $4.08 \pm 0.48 \text{ mg} \Gamma^1$. No significant difference was observed in DO values. pH value of ten ponds varied between 7.82 ± 0.31 in Pond-1 in the year 2009 (Site-1) to 8.08 ± 0.21 in Pond-2 of Site-2 in the year 2009. The average value of pH was found to be 7.89 ± 0.40 during 2009 and 2010. Salinity values were observed minimum of 20.66 ± 0.98 ppt in Pond-2 to maximum of 29.08 ± 1.62 ppt in Pond-1 of Site -1 in 2009. In both the sites, the average value of salinity in ten ponds was found to be 25.47 ± 2.99 ppt. Significant differences were observed in salinity values in different Ponds (F = 10.051, P ≤ 0.05). Transparency values were recorded in ten ponds having minimum of 24.00 ± 4.39 ppt in pond-2 of Site-2 in 2009 to maximum of 30.50 ± 5.72 ppt in pond-3 of Site-1 in 2009. Alkalinity values were ranged between $50 \text{ mg} \Gamma^1$ to $100 \text{ mg} \Gamma^1$ in both the years.

Minimum and maximum alkalinity was observed between 56.5 mg l^{-1} to 100 mg l^{-1} . Total hardness ranged from 100.2 mg l^{-1} in 2009 to 83.2 to 99.5 mg l^{-1} in 2010 where as variations among stations were very negligible.

Table 2 Correlation of *P.monodon* showing area of pond and diseased % (Site 1)

	Area(sqm)	Stocking Density	Survival	Affected
Area(sqm)	1.000			
Stocking Density	0.807**	1.000		
Survival	0.485**	0.864**	1.000	
Affected	0.715**	0.615**	0.313*	1.000

Table 3 Correlation coefficient of *P.monodon* showing area of pond and diseased % (Site 2)

	Area(sqm)	Stocking Density	Survival	Affected
Area(sqm)	1.000			
Stocking Density	0.836**	1.000		
Survival	0.303*	0.703**	1.000	
Affected	0.813**	0.527**	-0.154NS	1.000

No significant differences of salinity, transparency and hardness was found (F =10.051, 1.524 and 1.488 at P \leq 0.05). Pearson's correlation coefficient was calculated between various hydrological parameters and represented in Table 1.The DO showed negative correlation with temperature (r = -0.481 at P \leq 0.01), pH (r = -0.788), salinity (r = -0.014), and alkalinity (r = -0.749). Water transparency, hardness and calcium showed positive correlation (r = 0.690, 0.623, 0.652) with temperature (Table 1) at P \leq 0.01. Salinity was positively and significantly correlated with length and weight of the shrimp (r = 0.213, 0.235) where as Ca hardness was positively correlated with calcium (r = 0.679) at P \leq 0.01.

Correlation coefficient between area of ponds, stocking density, survival % and affected % was calculated which showed that stocking density was significantly correlated (r =0.807, p ≤ 0.01 & r = 0.836, p ≤ 0.01) with area of the pond (Table 2&3). Affected % showed significant correlation with area of the pond and stocking density (r =0.715, P ≤ 0.01 & r = 0.615, P ≤ 0.01) at site-1 and also showed positive (r =0.813, P ≤ 0.01 & r = 0.527, P ≤ 0.01) at site-2. Survival % was highly significant (r =0.485 at P ≤ 0.01) with area of the pond. Thus it was concluded that when stocking density was more, the more would be affected %(Fig.5 & 6). In site-2 survival % did not show any significance with affected %.

The mean affected % in 2010 was found to be higher in pond- 1 of site -2 whose area was less with high stocking density. The mean survival % in 2010 was found to be lower in pond-1 of site-2 whose area and stocking density was more (65000). In 2009, the % survival was less in pond-3, whose area was less and stocking density (60000) more (Fig. 7 & 8). Similarly the stocking density showed highly significant positive correlations with affected % in both the sites (F = 0.864, P \leq 0.01 & F = 0.527, P \leq 0.01). The highest % (22.67) of infected post larvae has been noticed in the month of June and lowest % (12.35) was observed in 30-45 days of culture in the month of April in both the years.



Fig.5 Affected % of P.monodon at Site 1 in 2009



Fig.6 Affected % of five cultured Ponds (Site2) in 2009-2010



Fig.7 Survival % of *P.monodon* in natural conditions at site 1



Fig.8 Survival % of *P. monodon* in natural conditions at site 2

DISCUSSION

The highest percentage (22.67%) infected post larvae were noticed in the month of June and lowest (12.35%) in April of the same year. The high survival rate of 92.17%, 91.53%, 91.21%, 90.78%, and 89.61% was observed in five ponds of Balasore in both the years. Water quality parameters were more or less similar. In the present study, the minimum dissolved oxygen was ranged above 4.0mgl^{-1} in both the years which were in agreement with the works of Bray et al., [8], Boyd [9]. Temperature is another factor which affect metabolism of shrimps in grow-out culture ponds. The average temperature should range between 25° C to 30° C and values greater than that may cause lethal effect in shrimps [9]. In the present study, the value of temperature was between 28° C to 30° C. Physiology of shrimps may be affected when salinity become less than 15 ppt or more than 25 ppt [10].

Further the stocking density of shrimps was one of the vital zoo- technical factors that influence the survival, growth, health and production yields [11]. The study showed significant affected % with high stocking density. Several studies confirmed that, increased stocking density cased negative effect on survival and growth of shrimps [11, 12]. The low oxygen level is a common problem in ponds with high stocking density that in turn increased the disease susceptibility. The black gill disease in prawns has been reported to accompany many other syndromes such as viral, bacterial, fungal and protozoan infections [13]. This disease in *P.monodon* takes the form of reddish, brownish to black gills. Adult *P.monodon* reared in ponds of northern Odisha (Balasore and Bhadrak district) were found to have black gills which were also confirmed earlier by works of Zeng [14] in Thai farm. Black gill disease was also confirmed by Lightener [15]. Examination of shrimps (by the respective farms) was revealed that black gill

disease might be a result of fungal and bacterial infection (*Fusarium spp. or zoothamniun*) as well as soil particles in the gills during heavy siltation [16].

CONCLUSION

Even with the available information on the diseases of *P.monodon*, many gaps still exist but could be bridged through a more intensive research programme. Infectious agents (microorganisms) were very difficult to identify. More effective methods of prevention and control are very much needed to combat not only fungal and protozoan diseases but bacterial, viral and non-infectious diseases as well.

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