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Comparative study of the reproductive performance and White Spot Syndrome Virus (WSSV) status of black tiger shrimp (*Penaeus monodon*) collected from the Bay of Bengal☆

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ABSTRACT

A comparative study to assess length, weight, fecundity, hatching rate and White Spot Syndrome Virus (WSSV) prevalence in black tiger shrimp (*Penaeus monodon*) broods collected from shallow and deep water zones of the Bay of Bengal was carried out in Cox's Bazar, Bangladesh. Average size and reproductive performance of broods from the deep zone were significantly higher than those of broods caught from the shallow zone. The incidence of WSSV infection in shallow zone broods was much higher than that in deep zone broods. The association between depth zone and WSSV infection is independent of brood size. WSSV infection is negatively associated with hatching rate, irrespective of location. Exclusive use of tiger shrimp broods collected from the deep zone could reduce vertical transmission of WSSV by 46% without additional management measures. A variety of economic, social and ecological factors make this potential solution unworkable however. A better alternative could be to promote the use of domesticated *P. monodon* broods.

1. Introduction

Shrimp farming in Bangladesh has expanded rapidly since the 1980s. The main species cultured is black tiger shrimp (*Penaeus monodon*), known as *bagda* in Bengali. At present, the area under tiger shrimp production is 188,046 ha, of which around 5% is located in the district of Cox's Bazar in the southeast of the country, and the remaining 95% in the districts of Khulna, Bagerhat and Satkhira in the southwest (Belton et al., 2011). Bangladesh produced 56,569 MT of tiger shrimp and 39,868 MT giant freshwater prawn (*Macrobrachium rosenbergii*) in 2010–2011 with an export value of approximately $462 million (DOF, 2012). The sector is the country's second largest source of export earnings after readymade garments, and shrimp farms alone directly employ in excess of 600,000 people (Karim et al., 2011; USAID, 2006). The majority of shrimp farms in the coastal region of Bangladesh follow extensive culture practices, relying mainly on natural productivity, with limited or no management in respect of drying and plowing the *gher*[1] bottom between crops, or liming, fertilization and feeding. The stocking density for PL typically ranges from 0.2 to 1.5 PL/m^2 and annual yields of shrimp are low; in the order of 160–230 kg/ha (Belton et al., 2011).

This low level of intensity is in part a risk mitigating strategy deployed by farmers in response to the pervasive and highly destructive White Spot Disease (WSD) (Karim et al., 2011). WSD, which is caused by White Spot Syndrome Virus (WSSV), is capable of causing 100% mortality within a few days of the onset of clinical signs (Ayub et al., 2008; Sanchez-Martinez et al., 2007; Vaseeharan et al., 2003). Until the recent emergence of EMS (Leano and Mohan, 2012), WSSV was considered the most serious disease problem for shrimp aquaculture in Asia (Hossain et al., 2001; Otta et al., 2003), where it over-shadowed 'all other disease agents as the leading cause of production losses' (Kanchanaphum et al., 1998, p1).

In Bangladesh, WSD first occurred in cultured tiger shrimp in semi-intensive farms in Cox's Bazar in 1994, resulting in their permanent closure. In 1996 the disease spread to Khulna region in the southwest of the country, affecting approximately 90% of extensive shrimp farms and causing a 20% drop in national shrimp production. Shrimp exports fell from 25,742 tonnes to 18,630 tonnes in 1997–1998. They subsequently rebounded in 1999–2000, only to fall by 25% in 2001 as production was again badly affected by WSSV associated with other viral and bacterial pathogens (Alam et al., 2007). WSD is now endemic and is generally regarded as one of the most important constraints to the industry's sustainability and further expansion in Bangladesh (Karim et al., 2011). As a result, finding ways to minimize the incidence of WSD could have significant implications for the future success of the industry.

The first shrimp hatchery in Bangladesh was established by the Department of Fisheries (DOF) at Cox's Bazar in 1987. At present, 57

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[1] *Gher* is the local term to the shallow embanked earthen enclosures in which shrimp are produced in Bangladesh.

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shrimp hatcheries are in operation, all of which are located in Cox's Bazar (BFFEA, 2009). Present annual demand of bagda PL, as reported by the secretary of the Shrimp Hatchery Association of Bangladesh (SHAB), is about 8 billion and present annual production is about 6–7 billion, where hatcheries have the capacity to produce 15 billion PL (Pers. Comm. Homayion, 2010). The gap between production and demand is met by harvest of wild PL.

Domesticated *P. monodon* broods are unavailable in Bangladesh and hatcheries are completely dependent on wild broods captured from the Bay of Bengal. However, unlike domesticated broodstock, which can be raised in a controlled, disease free environment, wild broodstock are frequently exposed to or infected by pathogens, including WSSV. WSSV can be vertically transmitted from WSSV-positive spawners to their offspring (Lo et al., 1996b). This has important implications for the prevention of disease at the growout stage since infected postlarvae can represent a major source of infection for shrimp farms (Sanchez-Martinez et al., 2007). This means that screening and selecting WSSV-negative brooders can markedly reduce the chances of a subsequent outbreak of WSSV (Vaseeharan et al., 2003), providing that appropriate management practices for reducing the likelihood of horizontal transmission during are also followed (Karim et al., 2011).

The presence of WSSV in wild caught *P. monodon* broods taken from hatcheries has been confirmed by a number of studies. Ayub et al. (2008) reported infection rates of 20% and 30% from samples of 60 spawners taken from hatcheries in Cox's Bazar district, using non-nested and more sensitive nested PCR testing techniques respectively. Research by Vaseeharan et al. (2003) revealed that 34% of *P. monodon* brooders taken from hatcheries on the east coast of India tested positive for WSSV by PCR, while Hossain et al. (2001) recorded the incidence of WSSV observed in Indian tiger shrimp broodstock at 50%, and Remany et al. (2012) found the prevalence of WSSV infection in tiger shrimp broods captured off the southeast coast of India to be 21%.

Lo et al. (1996b) also reported that approximately 48% of captured *P. monodon* broodstock tested in Taiwan over an eight month period were WSSV-positive, but that prevalence varied considerably with season. In another longitudinal study, Iqbal et al. (2011) found the incidence of WSSV in broods from hatcheries in Cox's Bazar to range from 0% in September to as high as 90% in May/June, with a similar, but delayed, pattern of temporal variation evident in nauplius and postlarvae. Withyachumnarnkul et al.'s detailed study from Thailand also revealed seasonal fluctuations in the percentage of WSSV-positive prevalence in *P. monodon* broods of between 0 and 18%, which were matched by a highly significantly correlated ($p \ll 0.01$) pattern of peaks and troughs in prevalence in PL occurring approximately 1 month after those for the broodstock (Withyachumnarnkul et al., 2003).[2]

The studies cited above clearly demonstrate the presence of WSSV in wild *P. monodon* broods used in many hatcheries, along with the likelihood of vertical transmission to the PL. However, none of them attempt to link the prevalence of infection to the origin of broods, with the exception of Vaseeharan et al. (2003) which only does so as far as the hatchery itself, and not to the fishery from which the spawners were captured.

One of the authors of this paper, working in the shrimp hatchery sector in Cox's Bazar for a number of years, has observed the commonly held perception among hatchery technicians that broods captured from deep zones of the Bay of Bengal are affected by WSSV less frequently than those from shallow zones. Discussion with owners of fishing vessels confirmed that they caught shrimp broods mainly from two separate depth zones; a deep zone and a shallow zone, where depths ranged from 40 to 50 m and 15 to 25 m respectively.

This paper attempts to evaluate the accuracy of the observation that *P. monodon* spawners harvested from shallow zones of the Bay of Bengal are more likely to test WSSV positive than those captured from deeper zones, and explores interactions between this factor and the seasonal variations in WSSV prevalence noted above. In order to achieve this, the present study sampled broods captured from shallow and deep zones of the Bay of Bengal from five hatcheries in Cox's Bazar over a six month period. WSSV was tested for using Polymerase Chain Reaction (PCR) testing facilities. Length, weight, fecundity and hatching success were also measured in order to check for any association with brood origin, or correlation with WSSV.

The paper also explores the possible implications of these findings for the shrimp sector, taking into account a number of social and economic features of hatcheries and brood supply networks which influence the likelihood of any corrective recommendations or policies being successfully implemented. In addition to quantitative data pertaining to shrimp broods, the Results section therefore also provides qualitative information regarding the characteristics of the brood fishery which are important to this analysis.

2. Material and methods

2.1. Sampling

The study was conducted in Cox's Bazar during March to August, 2010 (the peak season for production of shrimp PL). In each of the study months, data relating to 30 brood-mother shrimp was collected, of which 15 had been caught from the deep zone (40–50 m water depth) and 15 from the shallow zone (15–25 m water depth). Brood samples were collected from five shrimp hatcheries which were selected for their goodwill, cooperativeness, high standards and continuous production. As hatcheries purchase broods opportunistically according to their availability and the level of demand for PL (i.e. without reference to their place of origin in either the deep or shallow zones) it was not possible to guarantee a complete sample of shallow and deep zone broods ($15 + 15 = 30$) from any given hatchery in each month. However, by co-operating with five hatcheries it was always possible to find at least one hatchery with 15 broods originating from either deep or shallow zone in each month.

Each hatchery used approximately 250 to 350 broods for running a single production cycle (time from receipt of broods to sale of PL), with each cycle lasting 25 to 35 days (average = 30 days). A sample of 15 broods therefore represented approximately 5% of all the broods used in a cycle by a hatchery. In each case, the origin of broods (i.e. the depth zone from which they were captured) was ascertained by discussions with hatchery technicians who went to receive broods from fishing vessels. In three of the hatcheries, broods were held individually, or in pairs, in 500 l volume fiberglass tanks. In two hatcheries broods were held in larger tanks containing 20–30 individuals. In order to ensure that broods were sampled at random, in the first case one brood was selected sequentially from each 20th tank, and in the second case, a single animal was sampled from each brooder tank.

2.2. Size measurement (length, weight)

Length, weight, fecundity and the hatching rate of eggs were measured for each shrimp sampled. Shrimp body weight (g) and length (cm) were measured using a digital weighing machine (model: elb-300) and scale respectively.

2.3. Reproductive performance (fecundity, hatching rate)

Fecundity (i.e. total number of eggs found from one brood) was calculated by taking 10 samples from different parts of the spawning

[2] Shrimp may be WSSV-positive without succumbing to the virus. Onset of the disease often seems to be triggered or aggravated by exposure to stressors including rapidly changing environmental conditions which compromise immune status (Flegel and Alday-Sanz, 1998; Huang and Song, 1999; Kautsky et al., 2000). The delayed pattern of infection in PL results because the process of spawning brood and nursing nauplius to PL lasts approximately one month.