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Viral diseases of the giant fresh water prawn *Macrobrachium rosenbergii*: A review

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ABSTRACT

The giant freshwater prawn *Macrobrachium rosenbergii* is cultivated essentially in Southern and South-eastern Asian countries such as continental China, India, Thailand and Taiwan. To date, only two viral agents have been reported from this prawn. The first (HPV-type virus) was observed by chance 25 years ago in hypertrophied nuclei of hepatopancreatic epithelial cells and is closely related to members of the Parvoviridae family. The second, a nodavirus named *Mr*NV, is always associated with a non-autonomous satellite-like virus (XSV), and is the origin of so-called white tail disease (WTD) responsible for mass mortalities and important economic losses in hatcheries and farms for over a decade. After isolation and purification of these two particles, they were physico-chemically characterized and their genome sequenced. The *Mr*NV genome is formed with two single linear ss-RNA molecules, 3202 and 1250 nucleotides long, respectively. Each RNA segment contains only one ORF, ORF1 coding for the RNA-dependant RNA polymerase located on the long segment and ORF2 coding for the structural protein CP-43 located on the small one. The XSV genome (linear ss-RNA), 796 nucleotides long, contains a single ORF coding for the XSV coat protein CP-17. The XSV does not contain any RdRp gene and consequently needs the *Mr*NV polymerase to replicate.

Contents

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1. Introduction

The giant freshwater prawn, *Macrobrachium rosenbergii*, is an economically important crustacean, being farmed on a large scale in many different countries. Its culture is mostly developed in southern and South-Eastern Asian countries and to a lesser extent in the Caribbean (Northern South America and West Indies). In India its culture was introduced and developed approximately 8–10 years ago to help compensate for the substantial losses due to the epidemic white spot syndrome in marine shrimp (Penaeids) farming, hypothesizing the resistance of the giant freshwater prawn to white spot disease (WSD) (Sahul Hameed et al., 2000).

To date, only two viral diseases were reported in *M. rosenbergii*: the first one, affecting the digestive tract, is due to a parvo-like virus which was recorded for more than 18 years ago (Anderson et al., 1990); the second viral disease was reported during post-larvae mortalities (Arcier et al., 1999) and called white tail disease (WTD) due to the clinical signs observed, and was attributed to the simultaneous development of two different viral particles (Qian et al., 2003). Very few data are available for *M. rosenbergii* parvo-like virus. This review will therefore be focussed on WTD.

2. Parvo-like virus

Known as HPV-type (hepatopancreatic parvovirus-type) disease, because of the similarities of the histological symptoms recorded for the HPV disease reported in *Penaeus chinensis* (Lightner and Redman (1985) and more generally in penaeids, this disease is one of the very rare known diseases of *M. rosenbergii*. The pathogenic agent is a parvo-like virus that develops in nuclei of hepatopancreatic epithelial cells (Anderson et al., 1990). The infected nuclei become hypertrophied and exhibit a prominent basophilic intranuclear inclusion body surrounded by a clear area limited by the nuclear membrane associated with a thin chromatin layer. Using electron microscopy, the infected nuclei contain small icosahedral particles, about 29 nm in diameter. The pathogenic agent, by its size and location, is closely related to the members of the Parvoviridae. Moreover, symptoms reported look like those described for insect densovirus. This agent, strictly located in digestive tract of *M. rosenbergii* was only reported from Malaysia, and its prevalence and pathogenicity in wild and farmed populations of this freshwater prawn is unknown (Anderson et al., 1990).

Attempts to detect HPV-type of *M. rosenbergii* by *in situ* hybridization (ISH) using cloned probes of HPV isolated from *P. chinensis* were unsuccessful (Lightner et al., 1994). This clearly indicates important genome differences between these two viruses and consequently suggests that they are not or very remotely related. Further characterization of the pathogen is needed to confirm its identity and to better understand the resulting disease.

2.1. White tail disease

White tail disease (WTD) is responsible for large-scale mortalities in hatchery reared freshwater prawns, leading to subsequent economic losses. Though WTD is the most recently reported disease affecting *M. rosenbergii* (Arcier et al., 1999), it is also the one that has been the subject of the most investigations.

3. History

Since 1994, mortalities were episodically reported in a hatchery in Pointe Noire in Guadeloupe (French West Indies). These mortalities varied from 5% to 90% depending on the reproductive cycles of the brood stock. The first pathological sign noticed was the presence of whitish post-larvae (PLs) near the surface of the rearing tanks, followed by sudden mortalities starting just 3 days later. Initial investigations had hypothesized possible toxicity of the inlet water, or effects of chemicals issued from the building (i.e., paint toxicity, chlorine residues, tank osmosis, ground water pollution, nitrite contamination, anoxia, etc.). After having discarded all of these possibilities, investigations were carried out to look for the presence of a possible infectious agent (Herman, personal communication). Freshly dead PL's were collected and stored at −20 °C prior to being transferred (in 1997) to Montpellier laboratory for investigations.

4. Clinical signs and histopathology

The main sign gave the name to this disease: whitish coloration of muscles, starting in some areas of the tail, extending to the tail muscles (abdomen) and at a final stage to all the muscles of the prawn, comprising head (cephalothorax) muscles. These signs were associated with abnormal behavior, some lethargy and anorexia. Mortalities occurred after the first clinical signs (1–3 days) and all the PL population of a tank was often lost within less than one week (Herman, personal communication). When investigated by histology, lesions were evidenced essentially in muscle and connective tissues. They correspond to small dense basophilic inclusions, 0.5–3 μm in diameter, located in cytoplasm. Muscles were more or less necrotic and muscle fibers were dissociated. Epithelial tissues appeared normal, particularly the sub-cuticular and the digestive epithelium (Arcier et al., 1999).

5. Viral etiology of the disease

The viral aetiology of the disease was demonstrated very early by observing (via transmission electron microscopy (TEM) non-enveloped particles, about 27 nm in diameter, in tissue homogenates of diseased PL's. In parallel, in ultra-thin sections, similar sized particles were observed in the cytoplasm of connective cells in affected larvae. Due to its general characteristics, the causative agent of WTD was attributed to a nodavirus, named *M. rosenbergii* nodavirus (*Mr*NV) (Arcier et al., 1999). Later, a second virus-like particle, unusually small, 15 nm in diameter and consequently named XSV (extra small virus-like particles), was also found associated with *Mr*NV (Qian et al., 2003). They were both icosahedral in shape, unenveloped, and located in the cytoplasm of infected target cells (Qian et al., 2003). It is worthwhile to note that the two viruses were found in WTD-diseased animals collected in the French West Indies (Bonami and Sri Widada, unpublished data) as well as in diseased animals collected in China (Qian et al., 2003), in India (Tripathy et al., 2006) and in Thailand (Yoganandhan et al., 2006).

Transmission of the disease to healthy PLs by inoculation of infectious materials or immersion challenge caused 100% mortality but failed to cause mortality in adult prawns, even though virions were detected in tissues. The duration of the disease was dose dependant, but usually, cumulative mortality reached 100% after 8–14 days post-infection (Sahul Hameed et al., 2004a).

6. Geographic range

The disease was first reported in Guadeloupe in 1997 (Arcier et al., 1999), then in Martinique (French West Indies) and in Dominican Republic (Sri Widada and Bonami, unpublished data), in the People's Republic of China (Qian et al., 2003) and more recently, in India (Sahul Hameed et al., 2004a,b; Vijayan et al., 2005; Shekhar et al., 2006). A morphologically similar agent (*Mr*NV-type called MMV for *Macrobrachium* muscle virus) was observed in Taiwan (Tung et al., 1999). The resulting disease could be