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**ORIGINAL ARTICLE**

**Review Article**

# A Short Review on Infectious Viruses in Cultural Shrimps (*Penaeidae* Family).

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**Abstract:** A major constraint limiting the shrimp production is diseases. Shrimp aquaculture is an important industry in many countries especially Southeast Asia and Iran. In cultured pond, the shrimp may be infected with several pathogens such as several viruses. There are at least six lethal viruses affecting penaeid shrimps production in the world especially Southeast Asia and Thailand. However, known viral pathogen in shrimp is about 20. They have been identified from 1970. Incidence of infection in artificial condition is more than nature. The 6 viruses are very important and they cause serious problem for shrimp cultivation and economic losses. They are consisting of HPV, IHHNV, MBV, TSV, WSSV and YHV. Two of them are highly pathogenic and lethal in shrimp such as WSSV and TSV. Shrimp aquaculture is a successful activity. Despite this success, annual production decreased in the latter because of widespread epidemics (epizootics) caused by new viral pathogens. Molecular diagnostic methods such as PCR are tools to detection viral diseases in shrimp in many parts of the world. Pathological methods and electron microscopy are good tools to detection viral disease especially at the first outbreak. Sanitary methods are the best way to control and prevention of viral diseases.

**Keywords:** Shrimp; Viruses; Pathogen.

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# Introduction

Shrimp farming in the Asia-Pacific region is one of the most lucrative aquaculture sectors. Asia leads the world in cultivated shrimp production with export earnings in the order of billions of US dollars per year. The pioneers for shrimp cultivation have been Japan, India, Thailand, China, Philippines, Vietnam, Iran, Ecuador, Taiwan and some other countries in the Southeast of Asia and South America and so Australia. Thailand alone has been the world's leading producer since 1992 with its export earnings alone reaching more than 1 billion US dollars per year (1-3). Iran export shrimp. It is about 6ooo metric ton annually and its value is about 30 M$ per year. Iran not only cultivated shrimp but also is cultivating other aquatic animals such as Sturgeon, Rainbow trout, Carp and Tilapia.

Viruses are the most common biological agents in the marine environment and it is known that they infect Fish, Shrimp and other aquatic animals. Marine crustaceans can be simultaneously infected by more than one type of virus (4-6).

The major viruses of concern in shrimps and fresh water shrimp are mention in the following (1, 2, 7, 8):

1) White-spot syndrome virus (WSSV or PmNOBII a mistake name which called for WSSV).
2) Monodon baculovirus (MBV).
3) Yellow-head virus (YHV).
4) Hepatopancreatic parvovirus (HPV).
5) Related Australian lymphoid organ virus (LOV).
6) Gill associated virus (GAV).
7) Infectious hypodermal and hematopoeitic necrosis virus (IHHNV).
8) Taura syndrome virus (TSV).
9) Mourilyan virus (MOV).
10) Laem Singh virus (LSNV).
11) Baculovirus midgut gland necrosis virus (BMNV).
12) Monodon slow growth syndrome (MSGS).
13) Infectious myonecrosis virus (IMNV).
14) Macrobrachium rosenbergii nodavirus (MrNV).
15) Extra small virus (XSV).

More than 15 viruses have been reported to infect marine shrimp (1, 9). They cause disease in shrimp specially penaeid shrimp family as species as *Penaeus monodon*, *Litopenaeus vannamei*, *Fenneropenaeus indicus Litopenaeus stylirostris, Marsupenaeus japonicus* and etc (10-13). Nine viruses are responsible for main considerable economic losses. These include white spot syndrome virus (WSSV), infectious hypodermal and hematopoietic necrosis virus (IHHNV), monodon baculovirus (MBV), hepatopancreatic parvovirus (HPV), yellowhead virus (YHV), gill-associated virus (GAV), Taura syndrome virus (TSV), infectious myonecrosis virus (IMNV), and Mourilyan virus (MoV) (1). Although these viruses were no cause for alarm to human health, authorities find that they were economically crippling for Asian shrimp farmers (2).

Initially, *Penaeus monodon* was the main cultivated species in Asia but this has changed markedly since 2002 when *Litopenaeus vannamei* (formerly called *Penaeus vannamei*) started to be cultivated in many Asian countries. Since 2004, it has been the main cultivated species in the world (2).

Viral infection found not only in cultivated shrimp but also in wild shrimp. Different viruses have found in wild shrimps for example at a research which done in Brunei waters, Over 270 *P. monodon* were collected from the South China Sea, screened and spawned. Of the nine viruses assessed, infectious hypodermal and hematopoietic necrosis virus (IHHNV) was most commonly detected (19.6%), followed by monodon baculovirus (MBV) (7.4%), hepatopancreatic parvovirus (HPV) (3.8%), and Mourilyan virus (MoV) (0.9%). The only multiple viral infections found were a combination of IHHNV and MBV (2.2%). Two most infectious viruses for *P. monodon*, white spot syndrome virus (WSSV) and yellowhead virus (YHV) were not distinguished in any shrimp (1). Additional research in Thailand display number of viral infection in 42 shrimp samples from central and southern areas of Thailand using multiplex RT-PCR technique. Percentage of infection

in examined shrimp with different viruses was: HPV 4.8%, TSV 7.1%, YHV 2.4%, MBV 2.4%, IHHNV 2.4%, WSSV 40.5%, and Mix-infection 2.4% (14).

White Spot Syndrome Virus (WSSV) is the causative agent of widespread disease related with high mortality rate in cultured shrimp (12). It causes up to 100% mortality within 10 days in commercial shrimp farmhouses, resulting in huge losses to the shrimp farming industry (15). About 4–6 billion US$ of economic losses have been estimated in Asia and more than 1 billion US$ in America, between 1992 and 2001 and presently the disease has spread worldwide. Conventional control strategies such as improvement of environmental conditions, stocking of specific pathogen free (SPF) shrimp post-larvae and augmentation of disease resistance by oral immune stimulants, are currently employed to contain WSSV infections. However, extreme virulence of this virus and its wide host range including many other crustaceans make the transmission control and prevention to be problematic (16-21).

**Pathogenic DNA Viruses in Shrimp:**

DNA viruses which cause infection in shrimp contain: IHHNV, HPV, WSSV, MBV and BP.

**Pathogenic RNA Viruses in Shrimp:**

RNA viruses which are infectious for shrimp contain: YHV, GAV, LOV, TSV, IMNV, MOV, MrNV, XSV and LSNV.

Several positive sense RNA (+ssRNA) viruses have been reported from shrimp. Most notably, these include yellow head complex viruses (YHV) (22), and Taura syndrome virus (TSV) (6, 17, 23).

# Viruses

Properties of known viruses in shrimp summarized in below:

**Monodon baculovirus (MBV):**

**Introduction:** Even though MBV is not a severe pathogen for the black tiger prawn (*Penaeus monodon*), it should be eliminated from the farming system because it is unlikely that shrimp could carry such heavy viral infections without paying some price (5).

**Virion:** MBV is a DNA-virus like white spot syndrome virus (WSSV) and hepatopancreatic parvovirus (HPV). It is a double stranded DNA (dsDNA) virus (2, 14, 24).

**Signs:** MBV did not cause shrimp mortality so long as rearing conditions were good. The infection of this virus retards growth of the shrimp which named stunted-growth and resulted in economic losses (2, 14, 25-27).

**Diagnostic:** The viral inclusions can be seen directly through the cuticle of early PL specimens using the light microscope. In tissue sections stained with hematoxylin and eosin (H&E). Transmission electron microcopy is a potent tool to recognition viral particles in specimens. Polymerase chain reaction (PCR) is a rapid method for viral detection. However, experience has shown that the Taiwanese primers do not work with MBV from Thailand and Australia (2, 28, 29). Properties of the virus summarized in table 1.

**Yellow-head virus (YHV):**

**Introduction:** YHV was first mistakenly considered to be a baculovirus but it was soon discovered during purification and characterization that its morphology differed from that of baculoviruses (30, 31). Now, it is classified as *Ronivirdae* (32, 33).

**Virion:** Rod-shaped, enveloped virion. Its genome contain positive sense single-stranded RNA (+ssRNA) (20).

**Signs:** Gross signs of disease that included a yellowish cephalothorax and very pale overall coloration of moribund, infected shrimp (15).

**Table 1: Properties of MBV.**

| | | |
|---|---|---|
| **Name of virion:** | | **Monodon baculovirus (MBV).** |
| **Viral family:** | | ***Baculoviridae*** |
| **Name of disease:** | | Spherical baculovirusis (MBV-Disease). |
| **Host:** | | *Penaeus monodon*. |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** | Post larvae |
| | **(Shape** and **Genome)** | Icosahedral, Rod-shape,dsDNA, |
| **Epidemiology:** | **(Outbreaks)** | Mid 1980s Taiwan, 1990 Thailand, |
| **Virulence & Signs:** | | Acute, MBV did not cause mortality, but retards growth of the shrimp. |
| **Current Diagnosis Methods**: | | Pathological method (H&E Staining), Fluorescence microscopy, Monoclonal antibody, Immunohistochemistry, PCR, Nested PCR, Multiplex PCR, Dot-blot hybridization (Immuno dot-blot assay and Western blot test), in-situ hybridization, Scanning electron microscope, Transmission electron microscopy. |

**Diagnosis:** Histologically, YHV infections can be easily recognized by densely basophilic inclusions, particularly in H&E stained gill sections and rapidly stained whole gills, or by staining of hemolymph smears. Diagnosis is currently best by RT-PCR method rather than in situ hybridization. There is RT-PCR recognition kit based on the work done in Thailand and Australia. The kit is not useful to detect the types of YHV found in India. In addition to nucleic acid-based tests for YHV group viruses, monoclonal antibody assays have also been developed for diagnosis by immunohistochemistry. Dot blot assay and lateral flow chromatographic assay are accessible. Additional diagnostic rule is Gold-labeled MAb (Monoclonal Antibody) test strips (2, 19, 34-36)**.** The yellow head virus properties summarized in table 2.

**Table 2: Properties of YHV.**

| | | |
|---|---|---|
| **Name of virion:** | | **Yellow-head virus (YHV).** |
| **Viral family:** | | ***Ronivirdae*** |
| **Name of disease:** | | Yellow head disease (YHD). |
| **Host:** | | *Litopenaeus vannamei Litopenaeus stylirostris, Penaeus monodon,* *P. styliferus, Macrobrachium sintangense and M. lanchesteri.* |
| **Properties of Virion:** | **(Size, Shape, Enveloped** and **Genome)** | 40-50×150-200 nm, Rod shape (Bacilliform), Enveloped, +ssRNA. |
| **Epidemiology:** | **Reservoir & Vectors** | Invertebrates |
| | **(Outbreaks)** | 1990 and 1992 Thailand |

| | |
|---|---|
| **Virulence & Signs:** | Acute, YHV can cause high mortality in cultured shrimp. It is sometimes accompanied by the gross<br><br>signs of yellowing of the cephalothorax (from which the disease got its name) and general bleaching of body color. |
| **Current Diagnosis Methods:** | Histopathology of lymphoid organs and gills, PCR, *in-situ* hybridization, Monoclonal antibody assay, Immunohistochemistry (Dot-blot assay and Lateral flow chromatographic assay), Electron microscopy. |

**White-spot syndrome virus (WSSV):**

**Introduction:** Historically, WSSV was the second viral disease to seriously disturb Thai shrimp farmers. It has been demonstrated that spawning induces WSSV replication in *Penaeus monodon* (15, 37, 38).

**Virion:** This is a tailed, rod shaped, double stranded DNA virus with a very large circular genome in the order of 300 kbp. White spot syndrome virus belongs to the new virus family which called Nimaviridae and genus Whispovirus (2, 33, 39, 40). The G+C ratio of white spot syndrome virus (WSSV) is about 41% (41).

**Signs:** WSS-virus is highly pathogenic affecting various crustaceans. The infection in shrimp causes mortality up to 90–100% within 3–7 days post-infection. White spot disease (WSD) has been reported to cause severe mortality in farmed shrimp especially black tiger shrimp in many countries. Mass mortalities began to be reported with characteristic gross signs of WSSV infection (2, 14, 18, 42).

**Diagnosis:** Histopathology with the light and electron microscopes can indicate infection. On the basis of gross signs of disease, histopathology with the light and electron microscopes and molecular method based DNA methods used to distinguish virus. In situ DNA hybridization tests with cultivated shrimp of various species from several Asian countries that showed gross signs of white spot syndrome. PCR methods have been described for WSSV either singly (2). Methods for real-time PCR and isothermal DNA amplification have also been described. After development of DNA hybridization probes for WSSV in Thailand primers for PCR detection of WSSV were quickly developed. In addition to PCR tests, immunological tests have also been described, lateral flow chromatographic detection strips are now accessible from both Japan and Thailand (2, 14, 18, 42-44).

**Vaccine (Prevention and Control):** Trials show a new era for shrimp vaccination although shrimp immune system isn't like vertebrates. The results of some trials indicate the possibility of vaccination of kuruma shrimp with recombinant proteins against WSSV. However, new recombinant vaccine from VP28-Protein against WSSV is under studying. There is growing evidence that invertebrates, including crustaceans, possess some form of 'immune memory' or 'priming' mechanism that can be stimulated by past exposure to an infectious agent (24, 45-50). The properties of WSSV briefed in table 3.

**Table 3: Properties of WSSV.**

| | |
|---|---|
| **Name of virion:** | **White-spot syndrome virus (WSSV) or White spot virus (WSV).** |
| **Viral family:** | ***Nimaviridae*** |
| **Name of disease:** | White spot disease (WSD), HHNBV. |
| **Host:** | *Marsupenaeus japonicus, Penaeus* |

| | | |
|---|---|---|
| | | *monodon, P. penicillatus, P. semisulcatus, P. aztecus, Fenneropenaeus indicus, Litopenaeus vanname, Fenneropenaeus merguiensis, Fenneropenaeus chinensis, Farfantepenaeus duorarum, Litopenaeus schmittii, L. setiferus, L. stylirostris, Metapenaeus ensis, Macrobrachium rosenbergii.* |
| **Properties of Virion:** | **(Size, Shape, Enveloped** and **Genome)** | 80-120×250-380 nm, Rod shape to elliptical, with a tail-like extension, ovoid (bacilliform), Enveloped, dsDNA, Circular 290 to 305 Kbp. |
| **Epidemiology:** | **Reservoir & Vectors** | Invertebrates, Decapods (Crabs), Copepods, Crayfish, Aquatic insect larvae. |
| | **(Outbreaks)** | 1992-1993 Asia (southeast Asia and India), 1993 Japan, 1999 Central America, (Caused pandemic epizootic). |
| **Virulence & Signs:** | | Very Acute,<br><br>100% mortality within 3–10 days, shrimp with WSD are reported to show a rapid reduction in food consumption, become lethargic, have a loose cuticle with some showing characteristic white spots of 0.5–2.0 mm in diameter. |
| **Current Diagnosis Methods:** | | Histological methods (H&E Staining), Antibody based methods, Immunohistochemical tests, *in-situ* hybridization (*in-situ* DNA hybridization), Molecular methods (PCR, One-step PCR, nested PCR, real-time PCR) and Electron microscopy, Lateral flow chromatographic test. |
| **Appendix:** | | The largest of the known penaeid shrimp virus.<br><br>Previously called: Penaeid rod shape DNA virus (RDV), Rod shaped nuclear virus of *Ma.Japonicus* (RV-PJ), Hypodermal and hematopoietic necrosis baculovirus (HHNBV), White spot baculovirus (WSBV), PmNOBIII, Systemic ectodermal and mesodermal baculovirus (SEMBV) or PmNOBII, Chinese |

| | |
|---|---|
| | baculovirus (CBV). |

**Hepatopancreatic parvovirus (HPV):**

**Introduction:** HPV was first described from farmed marine shrimp in Singapore (51). Also, It is proposed to call this virus *P. merguiensis* densovirus (PmergDNV) (52, 53,

http://talk.ictvonline.org/media/p/380.aspx).
Hepatopancreatic parvovirus (HPV) is pathogen for cultivated and wild penaeid shrimp species including *Penaeus monodon* and *Fenneropenaeus chinensis* (11, 54, 55). Hepatopancreatic parvovirus infects the hepatopancreas in *Penaeus monodon* and is associated with slow growth that decreases profitability for shrimp farmers (25, 52).

**Virion:** HPV is a single-stranded DNA (ssDNA) virus that is non-enveloped icosahedral virus averaging 22–23 nm in diameter and containing linear ssDNA. As such it belongs in the family Parvoviridae in the densovirus group. Two Asian types have been characterized at the molecular level, one in *F. chinensis* from Korea and the other in *P. monodon* from Thailand. They differ in total genome length (approximately 4 and 5 kb, respectively) (2, 14, 44, 54, 56, 57). HPV genome consisted of 6321 nucleotides (54). HPV belongs to the Parvoviridae family, Parvoviruses are unique among all known viruses in having single-stranded DNA genomes which are linear. Virions are non-enveloped, containing a single copy of the small (4-6 kb) viral chromosome encapsidated

in a rugged icosahedral protein capsid 18-26 nm in diameter. (58-61). However, the classification of HPV viruses is still uncertain, because of their unusual capsid proteins and genome organizations (54, 58). There are dissimilar isolates for HPV. They are HPV-chinensis (HPVchin), HPVmonodon (PmDNV) and HPVsemisulcatus (HPVsemi) (53, 62).

**Epidemiology:** HPV may have been of Indo-Pacific origin but later spread to wild shrimp in the Americas via importation of live Asian shrimp for aquaculture. In any case, it is now considered worldwide in distribution. Horizontal and Vertical transmission of HPV have been described. It is proposed that HPV may have an unknown reservoir and carriers (2, 11).

**Signs:** The infection of this virus delays growth of the shrimp and resulted in economic losses (14, 27). Heavy infections caused in poor growth, which decreases shrimp production and without visible inflammatory response (2, 25, 54).

**Diagnosis:** Gross signs are not sufficient for HPV identification and other tests such as histological analysis or PCR testing are essential. DNA sequence analysis has revealed that there are different geographical kinds of HPV. It is recommended that different PCR primers be used for their detection (2, 54, 56). In addition to PCR, monoclonal antibodies for HPV detection have recently been produced (63) and it is hoped that a lateral flow chromatographic test will soon be available (2). Positive diagnosis is based on the presence of hepatopancreatic lesions showing basophilic inclusions within enlarged nuclei of tubule epithelial cells, and sometimes adjacent mid-gut cells (2, 11). Traditional PCR, PCR-ELISA, real-time PCR and histopathology for detection of shrimp hepatopancreatic parvovirus (PmDNV) is applicable. Loop-mediated isothermal amplification (LAMP) combined with amplicon detection by chromatographic lateral flowdipsticks (LFD) allowed simpler detection. In situ hybridization is another method to diagnosis (52, 54). A number of diagnostic methods were established for detection of this virus including histological method (H&E staining), Transmission electron microscopic (TEM), in situ hybridization, and polymerase chain reaction (PCR) (1, 2, 48, 64). Properties of the virus summarized in table 4.

**Table 4: Properties of HPV.**

| | |
|---|---|
| **Name of virion:** | **Hepatopancreatic parvovirus (HPV) or *Penaeus monodon* densovirus (PmDNV and PstDNV).** |
| **Viral family:** | ***Parvoviridae*** |

| | | |
|---|---|---|
| **Name of disease:** | | HPI (Hepatopancreatic parvovirus infection) |
| **Host:** | | *Penaeus monodon*, *Macrobrachium rosenbergii*, *Marsupenaeus japonicus, Fenneropenaeus merguiensis, Fenneropenaeus indicus, Fenneropenaeus chinensis, Penaeus orientalis.* |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** | Larvae, Post larvae and Juvenile. |
| | **(Size, Shape, Enveloped** and **Genome)** | 22-24 nm, Isometric (Icosahedral), Nonenveloped, -ssDNA, Linear, 4-5 Kbp, There are different geographical type of HPV. |
| **Epidemiology:** | **Reservoir & Vectors** | Vertebrates, Invertebrates |
| | **(Outbreaks, Transmission route)** | 1980s Singapore, Horizontal and Vertical. |
| **Virulence & Signs:** | | Slow growth that reduces profitability for shrimp farmers (reduced growth rates of prawns during the juvenile stages and overt mortalities). However, there are no specific gross signs for HPV so diagnosis may be difficult, High acute for larvae (The virus is lethal to shrimp larvae during the first month after stocking). |
| **Current Diagnosis Methods**: | | Histological examination (H&E staining), PCR and nested-PCR, PCR-ELISA detection method, The loop-mediated isothermal amplification (LAMP), Transmission electron microscopy (TEM), in-situ hybridization. |
| **Appendix:** | | The Australian HPV isolate from *P. merguiensis* is the fourth strain of penaeid prawn HPV to be partially sequenced.We have therefore proposed to name this virus *P. merguiensis* densovirus (PmergDNV), following the convention of the International Committee for the Taxonomy of Viruses. The other three are HPVchin from *P. chinensis* of Korea, PmDNV from P. monodon of Thailand and HPVsemi from *P. semisulcatus* of |

| | |
|---|---|
| | India. An additional strain of HPV has been reported in the freshwater prawn *M. rosenbergii.* |

**Infectious hypodermal and hematopoeitic necrosis virus (IHHNV):**

**Introduction:** The virus was first described in blue shrimp *Litopenaeus stylirostris* and white shrimp *L. vannamei* (Formerly called *Penaeus stylirostris* and *P. vannamei*) in the Americas in the early 1980s (2, 65, 66).

**Virion:** IHHN-virus is a non-enveloped, icosahedral virus with 22–23 nm in diameter and holding linear ssDNA of 4.1 kb. IHHNV is single-stranded DNA virus. Thus it is a typical densovirus classified in the Parvoviridae family (14, 44, 54, 58, 67, 68).

**Signs:** Although IHHNV is low virulence virus in adult shrimp (37) But, It causes acute epidemics and mass mortality only with juveniles and sub-adults of *L. stylirostris* (2, 67). The infection of this virus retards growth of shrimp so resulted in economic losses (14). In *L. vannamei*, it causes reduced, irregular growth and cuticular deformities that are collectively referred to as "runt-deformity syndrome" (RDS) (69-72). The infectious hypodermal and hematopoietic necrosis virus is very pathogenic for *Litopenaeus stylirostris* whereas infection in *Litopenaeus vannamei* is known to induce development and growth abnormalities and cause economic losses that range between 10% and 50% (12, 37).

**Diagnosis:** IHHNV can be identified by routine histological technique with H&E staining and in situ DNA hybridization assays with a specific IHHNV probe. A polymerase chain reaction (PCR) assay is also described in the OIE Aquatic Animal Health Manual. By TEM icosahedral virions can be seen in the cytoplasmic region of infected cells (2, 73, 74). Several clinical evaluations had shown that hemolymph is the best tissue sample for reliable diagnosis of IHHNV infection (37).

**Epdemiology:** In *L. stylirostris* the virus can transmit by vertical and horizontal route. IHHNV vertical transmission from infected females was clearly established (37, 67). Properties of this virus briefed in table 5.

**Table 5: Properties of IHHNV.**

| **Name of virion:** | | **Infectious hypodermal and hematopoeitic virus (IHHNV).** |
|---|---|---|
| **Viral family:** | | ***Parvoviridae*** |
| **Name of disease:** | | Infectious hypodermal and hematopoietic necrosis (IHHN) OR: Runt-deformity syndrome (RDS). |
| **Host:** | | *Litopenaeus stylirostri,and Litopenaeus vannamei.* |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** | Juvenile and Subadult. |
| | **(Size, Shape, Enveloped** and **Genome)** | 22-23 nm, Icosahedral, Nonenveloped, ssDNA, Linear,Lengh: 4.1 Kb,<br>Variants: IHHNV-I, IHHNV-II, IHHNVIII |
| **Epidemiology:** | **Reservoir & Vectors** | Shrimp |
| | **(Outbreaks, Transmission route)** | 1980s Taiwan & 1981 America, Pass onto other population by |

| | | |
|---|---|---|
| | | Horizontal & Vertical transmission. |
| **Virulence & Signs:** | | Acute epizootics and mass mortalities (>90%) in *L.stylirostris,* (In *L.vannamei,* It cause reduced-irregular growth and cuticular deformities on the other hand reported that it is asymptomatic without significant mortalities in *L.vannamei*) |
| **Current Diagnosis Methods:** | | It cause reduced-irregular growth and cuticular deformities on the other hand reported that it is asymptomatic without significant mortalities in *L.vannamei*) Histopathological staining and examination, in-sito DNA hybridization, Dot-Blot test, PCR, real-time PCR. |
| **Appendix:** | | The smallest of the known penaeid shrimp viruses. |

**Taura syndrome virus (TSV):**

**Introduction:** Taura syndrome was first known as a new disease in the Americas in 1992 but its viral etiology was not established until 1994 (75-77). It is a recent viral pathogenic agent to arrive on the Asian scene (11, 78, 79).

**Virion:** It is a naked (without envelop) 32 nm icosahedral virus containing ssRNA molecule with 10.2 kb length and positive sense (80). However, it was later allocated to the family Dicistroviridae near the genus Cripavirus (cricket paralysis virus) (32, 81). GC content of the viral RNA ranging from 35 to 45%. RNA constitutes about 30% of the virion weight. A small genome-linked virus protein (VPg), is covalently attached to the 5' end of the genome (41).

**Signs:** Experimental bioassays have exposed that low-grade mortalities can occur and that *P. monodon* can carry asymptomatic infections (1, 82, 83). Properties of TSV summarized in table 6.

**Table 6: Properties of TSV.**

| **Name of virion:** | **Taura syndrome virus (TSV).** |
|---|---|
| **Viral Family:** | ***Dicistroviridae*** (Previously classified as: *Picorniviridae*). |
| **Name of disease:** | Taura syndrome disease (TSD). |
| **Host:** | *Litopenaeus vannamei, Litopenaeus stylirostris, L.setiferus, L.schmitti.* Other penaeid *(Farfantepenaeus aztecus, Fa.duorarum, Fenneropenaeus chinensis, Penaeus monodon and Marsupenaeus japonicus)* have been experimentally infected. *Penaeus* |

| | | |
|---|---|---|
| | | *monodon*. |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** | Post larvae, Juvenile and Sub adult. |
| | **(Size, Shape, Enveloped** and **Genome)** | 32 nm, Isometric (Icosahedral), Nonenveloped, +ssRNA, 10.2 Kb. |
| **Epidemiology:** | **Reservoir & Vectors** | Invertebrates, Shrimp eating birds and Flying aquatic insects. |
| | **(Outbreaks, Transmission route)** | 1991-1992 Ecuador & 1999 Asia (1998 Taiwan), Horizontal and Vertical, Cause pandemic epizootic. |
| **Virulence & Signs:** | | Acute, Cumulative mortalities due to TSV epizootics have ranged from 40 to >90% in cultured population of *L.vannamei*. |
| **Current Diagnosis Methods:** | | Pathological methods (H&E staining), Monoclonal antibody based methods, in-sito hybridization, Dot-blot method (Western blot), Immunohistochemistry methods, DNA amplification method (PCR). |

**Mourilyan virus (MoV):**

**Introduction:** This virus was discovered accidentally during the study of GAV, an Australian virus from the yellow head virus complex (84).

**Epidemiology:** It appears to be endemic in populations of *Penaeus monodon* from Queensland in Australia, from Malaysia and from Thailand (2).

**Disease:** There is some indication that it may be associated with gradual, progressive mortality in pond-reared *Marsupenaeus japonicus* in Australia (85).

**Virion:** MoV size is about 85-100 nm in diameter. It is an enveloped virus with spherical to ovoid virions. The MoV genome is contained of single-stranded, negative sense RNA divided into 4 fragments. The morphology, genome type, genome fragmentation and genome organization most closely look like features of viruses in the family Bunyaviridaen (2).

**Diagnosis:** A nested RT-PCR method has been developed (86).

Table 7 shows the properties of MoV.

**Table 7: Properties of MOV.**

| | |
|---|---|
| **Name of virion:** | **Mourilyan virus (MOV).** |
| **Viral family:** | **The morphology, genome type, genome fragmentation and genome organization most closely resemble features of viruses in the family Bunyaviridae.** |
| **Host:** | *Penaeus monodon, Marsupenaeus japonica.* |

| | | |
|---|---|---|
| **Properties of Virion:** | **(Size, Shape, Enveloped** and **Genome)** | 85-100 nm, Spherical (ovoid), Enveloped, -ssRNA (4 Fragments). |
| **Epidemiology:** | **(Outbreaks)** | Australia, Malaysia, Thailand. |
| **Virulence & Signs:** | | There is some indication that it may be associated with gradual, progressive mortality in pond-reared *P. japonicus* in Australia. The link to specific disease in *P. monodon* is less clear. |
| **Current Diagnosis Methods:** | | Nested RT-PCR |
| **Appendix:** | | This virus was discovered accidentally during the study of GAV. |

**Laem Singh virus (LSNV):**

**Introduction:** Laem Singh virus (LSNV) is a new shrimp virus from Thailand (6).

**Virion:** This virus hasn't envelopment. Its shape is icosahedral which size is about 27 nm diameter, similar to the size of viruses in the family Luteoviridae. On the other hand, LSNV is like family Barnaviridae based on some its properties (2).

**Diagnosis:** In situ hybridization test and RT-PCR apply to detect infected shrimp (2). Features of LSNV summarized in table 8.

**Table 8: Properties of LSNV.**

| | | |
|---|---|---|
| **Name of virion:** | | **Laem Singh virus (LSNV).** |
| **Viral family:** | | **It is mostly from the family *Luteoviridae.* but also to mushroom bacilliform virus *(Luteoviridae or Barnaviridae)*.** |
| **Name of disease:** | | Monodon slow growth syndrome (MSGS) |
| **Host:** | | *Penaeus monodon*. |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** **(Size, Shape, Enveloped** and **Genome)** | 27 nm, Icosahedral, Nonenveloped, More genome information will be needed for proper classification of this virus. |
| **Epidemiology:** | **(Outbreaks)** | 2002 Asia. |
| **Virulence & Signs:** | | Probably, It cause monodon slow growth syndrome (MSGS) but more research will be needed. |
| **Current Diagnosis Methods:** | | *In-situ* hybridization, RT-PCR, Electron microscopy. |
| **Appendix:** | | Tests using both in situ hybridization and RT-PCR revealed the presence of |

| | |
|---|---|
| | LSNVin both MSGS ponds and normal growth ponds, indicating that it was probably not the direct cause of MSGS. There still remains the possibility the MSGS is related to the prevalence or severity of MSGS infections in a shrimp culture pond. |

**Baculovirus midgut gland necrosis virus (BMNV):**

**Introduction:** BMNV was acute pathogen of larval stages of *M. japonicus* in Japan in the early period of shrimp culture development but was excluded from the cultivation system after the mode of transmission from infected broodstock was established, and thorough washing of the eggs or nauplii was employed as a routine preventative measure (87). Characteristics of BMNV summarized in table 9.

**Table 9: Properties of BMNV.**

| | | |
|---|---|---|
| **Name of virion:** | | **Baculovirus midgut gland necrosis virus** |
| **Viral family:** | | ***Baculoviridae*** |
| **Name of disease:** | | BMN (It have five names more). |
| **Host:** | | *Marsupenaeus japonicus, Penaeus monodon.* |
| **Properties of Virion:** | **(Size)** | 36×250 nm |
| **Virulence & Signs:** | | Acute |

**Monodon slow growth syndrome (MSGS caused by Laem Singh virus (LSNV)):**

**Introduction:** Monodon slow growth syndrome (MSGS) was first observed by shrimp farmers in cultured black tiger shrimp in 2002 (6, 88). It may be a viral disease of shrimp. Several scientists acclaim that Laem Singh virus **(**LSNV) is an agent that cause MSGS (6).

**Signs:** Unusual slow growth in cultivated *P. monodon.* Properties of LSN-virus summarized in table 10.

**Table 10: Properties of LSNV which cause MSGS.**

| | | |
|---|---|---|
| **Name of virion:** | | **Laem Singh virus (LSNV).** |
| **Viral family:** | | **It is mostly from the family *Luteoviridae*. but also to mushroom bacilliform virus *(Luteoviridae or Barnaviridae)*.** |
| **Name of disease:** | | Monodon slow growth syndrome (MSGS) |
| **Host:** | | *Penaeus monodon*. |
| **Properties of Virion:** | **(Size, Shape, Enveloped** and **Genome)** | 27 nm, Icosahedral, Nonenveloped, More genome information will be needed for proper classification of this virus. |
| **Epidemiology:** | **(Outbreak)** | 2002 Asia. |
| **Virulence & Signs:** | | Probably, It cause monodon<br><br>slow growth syndrome (MSGS) but more research |

| | |
|---|---|
| | will be needed. |
| **Current Diagnosis Methods:** | *In-situ* hybridization, RT-PCR, Electron microscopy. |
| **Appendix:** | Tests using both in situ hybridization and RT-PCR revealed the presence of LSNVin both MSGS ponds and normal growth ponds, indicating that it was probably not the direct cause of MSGS. There still remains the possibility the MSGS is related to the prevalence or severity of MSGS infections in a shrimp culture pond. |

**Infectious myonecrosis virus (IMNV):**

**Introduction:** IMNV is the most recent of the identified shrimp viruses to arrive in Asia (89), and is thought to have been introduced with contaminated *L. vannamei* stocks from Brazil (1; 90). While *L. vannamei* is the primary host for IMNV, other species such as *P. monodon* are vulnerable by experimental infection (1, 91).

**Signs:** No gross signs or mortalities have been reported (1). Features of the virus summarized in table 11.

**Table 11: Properties of IMNV.**

| | | |
|---|---|---|
| **Name of virion:** | | **Infectious myonecrosis virus (IMNV).** |
| **Viral family:** | | ***Totiviridae* (A toti-like virus)** |
| **Name of disease:** | | Infectious myonecrosis (IMN). |
| **Host:** | | *Litopenaeus vannamei*. |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** | Juvenile and sub adult. |
| | **(Size, Shape** and **Genome)** | 40 nm, Icosahedral, A single dsRNA with 7560 bp. |
| **Epidemiology:** | **(Outbreaks)** | 2004 America & 2006 Asia, |
| **Virulence & Signs:** | | IMN presents as a disease in *L. vannamei* with an acute onset of gross signs and elevated mortalities, but it progresses with a more chronic course accompanied by persistent moderate mortalities. |
| **Current Diagnosis Methods:** | | Histological examination (H&E staining), PCR (One step PCR, Nested PCR, real-time PCR), in-situ |

| | |
|---|---|
| | hybridization. |

**Gill associated virus (GAV):**

**Introduction:** Gill-associated virus (GAV) is a RNA virus. In Australia, Gill-associated virus has been linked to morbidity and mortalities in cultivated *Penaeus monodon* (92-95). GAV also can to cause disease and mortalities similar to that caused by the more highly virulent Yellow-head virus (YHV) that continues to cause production losses in shrimp farmed in South-east Asia. GAV has a 26.2 kb ssRNA genome and is the type species of the Okavirus genus in the Roniviridae (92). Table 12 show properties of gill-associated virus.

**Table 12: Properties of GAV.**

| | | |
|---|---|---|
| **Name of virion:** | | **Gill associated virus (GAV), GAV is the australian strain of YHV.** |
| **Viral family:** | | ***Ronivirdae*** |
| **Name of disease:** | | Yellow head disease (YHD). |
| **Host:** | | *Penaeus monodon*. |
| **Properties of Virion:** | **(Genome)** | +ssRNA, 26.2 Kb. |
| **Current Diagnosis Methods:** | | See: YHV details. |
| **Appendix:** | | LOV and GAV share approximately 95% DNA sequence identity and 100% amino acid identity, establishing that<br><br>they are the same virus type, while GAV and YHV share approximately 85% DNA sequence identity and 96% amino acid identity indicating that they are different types. |

**Macrobrachium rosenbergii nodavirus (MrNV):**

*Macrobrachium rosenbergii* is fresh water shrimp. White tail disease (WTD) caused by both *Macrobrachium rosenbergii*-nodavirus (MrNV) and extra small viruses (XSV). MrNV is a satellite virus particles are found in *Macrobrachium rosenbergii* (giant river prawn) infected with Macrobrachium rosenbergii nodavirus (MrNV; a virus not yet classified but clearly related to viruses in the family *Nodaviridae*) (41). WTD is a major problem. It is responsible for severe mortality in post-larvae of *M.rosenbergii* in the hatcheries and nurseries (8, 96). White tail disease has been observed in freshwater prawn hatcheries and nursery ponds in different parts of India, causing high mortalities and huge economic losses (96, 97).

**Virion:** MrNV is a small, icosahedral, non-enveloped virus. Its size is 26–27 nm in diameter. The genome of MrNV composed of two pieces of ssRNA (RNA1 and RNA2) of 2.9 and 1.26kb, respectively, and there is a single polypeptide of 43kDa in the capsid (8, 98).

**Signs:** MrNV cause white tail disease (WTD) of freshwater prawns (8).

**Diagnosis:** A number of diagnostic methods have been established to identify this virus including histopathology, immunological methods, reverse transcriptase-polymerase chain reaction technique (RT-PCR) and in-situ dot blot hybridization method using nucleic acid probes. A sandwich enzyme-linked immunosorbent assay have been developed to detect MrNV in freshwater prawns (99). Recently, genome-based methods, dot-blot hybridization and RT-PCR have been developed to detect MrNV (8, 100).

**Epidemiology:** White tail disease was first described in the French West Indies (101), later in China (98),

India (97), then in Thailand (102) and recently in Taiwan (103). Characteristics of the virus summarized in table 13.

| | | |
|---|---|---|
| **Name of virion:** | | ***Macrobrachium rosenbergii* nodavirus (MrNV).** |
| **Viraln family:** | | ***Nodaviridae*** |
| **Name of disease:** | | White tail disease (WTD). |
| **Host:** | | *Macrobrachium rosenbergii* (Freshwater prawns). |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** | Larvae and Postlarvae. |
| | **(Size, Shape, Enveloped** and **Genome)** | 26–27 nm, Icosahehral, Nonenveloped, ssRNA, (2 pieces , 2.9 and 1.26kb). |
| **Epidemiology:** | **Reservoir & Vectors** | It is possibe of the marine shrimp (*Fenneroenaeus indicus, Marsupenaeus japonicus and Penaeus monodon*)<br>acting as reservoir for MrNV. |
| | **(Outbreaks)** | French West Indies, Taiwan, China, India. |
| **Virulence & Signs:** | | Causing high mortalities and<br>huge economic losses in hatcheries and nursery<br>ponds. |
| **Current Diagnosis Methods:** | | Histopathology, immunological<br>methods, reverse transcriptase-polymerase chain<br>reaction technique (RT-PCR) and in-situ dot blot hybridization method. |
| **Appendix:** | | The results of a study indicate the possibility of marine shrimp acting as reservoir for MrNV and XSV and maintaining their virulence in tissue system of marine shrimp. |

**Extra small virus (XSV):**

**Introduction:** The XSV (extra small virus) is a satellite virus particles are about 15 nm in diameter and serologically unrelated to those of MrNV. XSV is a positive-sense single-stranded RNA, about 800 bases in size, encoding a 17 kDa capsid protein. The mixed infection of MrNV and XSV is implicated in white spot disease of prawns. Extra small virus (XSV) may cause disease of shrimp and may produce white tail disease (WTD). It is may responsible for mortality in post-larvae of *M. rosenbergii* in the hatcheries and nurseries (8, 96). It has reported the presence of XSV in addition to MrNV in WTD-infected postlarvae of freshwater prawns in India. XSV does not cause mortality in marine shrimp as observed in adult freshwater prawn (8, 104).

**Virion:** XSV is a virus-like particle (A satellite virus particles), icosahedral in shape and 15 nm in diameter, with a linear ssRNA (98).

**Diagnosis:** Various methods have been developed to detect this virus including: histopathology, immunological methods, reverse transcriptase-polymerase chain reaction technique (RT-PCR), in-situ dot blot hybridization method, sandwich enzyme-linked immunosorbent assay, genome-based methods, dot-blot hybridization and RT-PCR (8, 97, 100, 104). Table 14 show properties of the virus.

**Table 14: Properties of XSV.**

| | | |
|---|---|---|
| **Name of virion:** | | **Extra small virus (XSV).** |
| **Viral family:** | | **No classified.** It is a satellite virus particles. |
| **Name of disease:** | | White tail disease (WTD). |
| **Host:** | | *Macrobrachium rosenbergii* (Freshwater prawns). |
| **Properties of Virion:** | **Sensitive growth stage of shrimp.** | Larvae and Postlarvae. |
| | **(Size** and **Genome)** | 15 nm, ssRNA, Linear. |
| **Epidemiology:** | **Reservoir & Vectors** | It is possibe of the marine shrimp (*Fenneroenaeus indicus, Marsupenaeus japonicus and Penaeus monodon*) acting as reservoir for XSV. |
| | **(Outbreaks)** | China & India. |
| **Current Diagnosis Methods:** | | Histopathology, immunological methods, reverse transcriptase-polymerase chain reaction technique (RT-PCR) and in-situ dot blot hybridization method. |
| **Appendix:** | | Have reported the presence of XSV in addition to MrNV in WTD-infected postlarvae of freshwater prawns in India. |

**Miscellaneous other viruses:**

A number of other viruses have been reported from cultivated shrimp in Asia and have been adequately covered in previous reviews. However, with the exception of baculovirus midgut gland necrosis virus (BMNV), none have been reported to be the cause of serious or widespread economic losses, and they are not covered in this review.

# Result and Conclusion

It is about 20 viruses which have found in shrimps from 1980 to 2011 as well as pathogens. On the other words, a new type of viral pathogen found in shrimp every 1-2 years. Shrimp cultivation is going to progress more and more in several countries and scientists have been found new viruses more, every several years. The most important viral pathogens in shrimp are WSSV, YHV, MBV, TSV, IHHNV and HPV which some of them cause severe mortality in ponds.

There is not report with human illness by viral disease in shrimps, but it is need more research.

Viruses of shrimps often cause no gross signs of disease in shrimp, especially in the natural environment. In stressful environments such as culture systems, some of these viruses can become more virulent and cause significant economic loss by mortality or retarded growth. But some of them are really lethal such as WSSV (4,13).

Viruses outbreak which had been seen and reported are contain: 1992 Thailand (HPV), 1995 Thailand (YHV), 1996-7 Thailand (WSSV), 1993 Japan and China (WSSV), 1980s Taiwan (MBV), 1990 Thailand (MBV), 1984 Singapore (HPV), 2003 India (HPV), 1980s America (IHHNV), 1992 America (TSV), 2002 Thailand (MSGS), 1999 French West Indies (WT-Disease). In some cases it is possible the ponds be infected with 2 or more viruses simultaneously. Shrimp feeding behavior specially cannibalism may also be a serious problem for shrimp farmers because it caused horizontal transmission of viruses (2).

Methods which applied to detection viral disease in shrimp is different they are listed in the following:

1. Histology (H&E staining- Light Microscopy).
2. TEM (Transmission electron microcopy).
3. Non-nested PCR.
4. Nested PCR.
5. Multiplex PCR.
6. Multiplex reverse transcription-polymerase chain reaction (mRT-PCR).
7. Real-time RT-PCR.
8. Multiplex RT-nested PCR.
9. Miniarray.
10. Single-step multiplex PCR.
11. Single PCR.
12. ELISA (Monoclonal antibody assay based test).
13. PCR–ELISA.
14. Fluorescence microscopy (Eosin formula used contains some phloxine dye to detect occlusion bodies).
15. *In situ* hybridization (A type of Nucleic acid-based test).
16. Monoclonal antibody assay based tests: (ELISA, Dot blot assay, Lateral flow chromatographic assay).

The best way to control and prevention of viral diseases is following of hygienic rules in breeding, nursery and ponds as well as sanitary methods. Application and improvement of SPF stocks is another way. Also, stocking and Cultivation SPR shrimp may be useful. Although immune system in shrimp is primitive but vaccination may be useful method for prevention of viral disease in future.

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