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Taura Syndrome Virus (TSV)

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Shrimp get sick too. Taura syndrome is an infectious disease caused by the Taura syndrome virus (TSV). TSV infects tissues of ectodermal and mesodermal origin including whole body hypodermis (cuticular epithelium) and gills. However, endoderm-derived organs such as the hepatopancreas, midgut, midgut caeca, muscle and heart, do not show histological evidence of TSV.

Taura syndrome was first reported in *Penaeus vannamei* from the Taura region in Ecuador in 1997 and then promptly spread to other American countries and other continents including Asia. Ecuadorian shrimp farmers first believed the disease was caused by the use of pesticides in banana plantations close to shrimp farms, however, scientific research confirmed it a viral etiology. TSV is a reportable OIE disease.

TSV is a very small virion (32 nm) and its genome consists of a linear, positive-sense single-stranded RNA that replicates in the cytoplasm of the target cells. It is suspected that TSV causes increased health problems in densely populated aquatic animal production environments. Taura syndrome disease may occur more often when salinity is below 30 ppt. Although only a single disease, severity may vary based on the virus strains infecting shrimp. Accordingly, 4 different TSV genotypes have been reported: group 1: Americas, group 2: South-East Asia, group 3: Belize, and group 4: Venezuelan.

Importantly, TSV infection is characterized by mass mortality (40 to over 90%) and is observed at the early stage of juvenile farming, 14-days after pond stocking. TSV can be spread between pond populations through horizontal transmission (cannibalism) and virus detection in early postlarvae suggests that vertical transmission is also possible. TSV infection can be detected by PCR analysis in almost all shrimp life stages including postlarvae, juveniles and adults of *P. vannamei* but not in eggs, larvae or zygotes.

Causative agent of Taura syndrome. The pathogenic agent Taura syndrome virus (TSV) belongs to the Genus Aparavirus, Family Dicistroviridae, Order Picornavirales. The disease was first characterized and reported in Ecuador during 1997 from farmed *P. vannamei*. TSV can be transmitted to susceptible shrimp populations by vectors that include infected feces of sea gulls (Larus atricilla), chicken (Gallus gallus) and aquatic insects like water boatman (Trichocorixa reticulate).

Susceptible species for TSV infection in which viral presence has been demonstrated and disease with clinical signs has been observed, include *P. monodon*, *P. vannamei*, *P. stylirostris*, *P. setiferus*, *P. aztecus*, and *Metapenaeus ensis*. Other species of aquatic organisms have been reported to be TSV PCR positive but not having active infection.

Clinical signs of TSV. The appearance of clinical signs can occur as early as late postlarvae or juvenile stages in nursery or grow-out ponds. Three disease phases have been differentiated.

Acute phase: Moribund shrimp due to TSV are observed during this phase. Sick animals display red chromatophores expansion, general pale reddish discoloration, uropods and pleopods distinctly red, soft shell, empty gut and are often sick in the late D stage of the moult cycle (post-moult). Severely affected shrimp die during C-D stage of molting (ecdysis). Only during acute phase it is possible to observe pathognomonic histological findings characterized by multifocal necrosis in cuticular epithelium of body surface, gills, appendages, hindgut, esophagus, and anterior and posterior stomach chambers. The antennal gland can also be destroyed in severely affected organisms due to TSV infection.





In addition to histological diagnosis, wet mounts could be useful to demonstrate TSV in sick shrimp during the acute phase by observing under light microscopy abundant spherical dark structures which are necrotic cells, characterized by presence of pyknotic and karyorrhectic nuclei and cytoplasmic remnants.

Recovery phase: Survivor shrimp from the acute phase then enter a recovery phase. Shrimp in this phase can be observed, while others may remain in the acute phase and daily mortality can be present in the pond. Common clinical signs during the recovery phase include dark-black or dark-brown- randomly placed multifocal and irregularly shaped cuticular melanized lesions. These are sites of resolving TSV lesions in the cuticular epithelium. Sick animals in this phase may have soft shells and generalized expansion of red chromatophores. Their feeding and behavior usually look normal.

Chronic phase: When shrimp recuperate from recovery phase and cuticular melanized lesions disappear by successfully moulting, they establish a new healthy exoskeleton with no evidence of TSV disease. Survivor shrimp may stay infected throughout their life with no clinical signs but being less resistant to environmental stressors present in the farming environment. TSV can be detected in lymphoid organs during the chronic phase by PCR.

Early detection using Shrimp MultiPath™. Early testing and detection with SMP can give farmers two to six weeks' notice before clinical signs appear and prior to mass mortalities. In commercial nursery ponds and grow-out shrimp ponds, TSV infection can be detected early, and farmers advised as soon as juveniles are stocked in ponds. This information is an early warning system preparing farmers for a critical period when slowing the spread of the disease and maximizing production outputs is still possible.

Early detection empowers the implementation of prompt mitigation strategies. These can include:

- PCR assays for pre-screening of broodstock before placing in production tanks
- PCR assays for pre-screening of postlarval shrimp discarding tanks that test positive for TSV infection
- Suspending pond stocking with postlarvae from infected hatcheries
- Avoiding live and fresh feeds (especially for broodstock) from countries with historic status of TSV infections
- Not feeding female broodstock 6 hours before moving to spawning tanks to reduce eggs contamination
 with feces, and reinforcing egg and nauplii washing and disinfection before transferring to hatchery tanks
 to reduce possible TSV contamination from broodstock feces
- Use postlarvae from breeding programs focused on exclusion plans and production of TSV-Free or Specific Pathogen Resistant (SPF/SPR) TSV-resistant or tolerant postlarvae
- Pond stocking only with TSV PCR tested postlarvae and, pond frequent surveillance for TSV using molecular tools, are procedures that will help control TSV infections

Preventive farming strategies that may reduce TSV transmission include:

- Restocking of entire farming zones with TSV-free stocks
- Removing sick or dead shrimp to prevent transmission through cannibalism
- Reducing pond density (partial harvest)
- Organic debris and feces removal (syphoning and/or bacterial bioremediation when possible), must be considered as priority tasks
- Proper technical assistance for periodic monitoring with appropriate diagnostic tools will allow for discrimination between TSV and other disease with similar clinical signs
- Biosecurity around infected ponds must be increased, for example management of affected ponds in daily routines, separating nets and equipment, physical barriers put in place, inform adjacent farmers of the infection, and harvest when commercial size is reached. Disease mitigation plans should include pathogen exclusion programs



Shrimp MultiPath^{impsi} is used to confirm when broodstock or postlarvae are infected with TSV infective particles. This data can be used to eliminate infected broodstock and/or postlarvae batches from production systems before stocking ponds with infected organisms.

Target life-history stages for accurate early detection include late postlarvae stages (both at hatchery and/or at farm raceways and nursery ponds), juveniles and adults. No evidence of TSV infection in zygotes, eggs or larvae has been documented. However, it is plausible the TSV might be transmitted vertically from adult to egg.

Target organs for sensitive Shrimp **Multi**Path[™] detection are pleopods, haemolymph and whole small shrimp. Adequate tissue sampling is essential for accuracy of TSV molecular detection and quantitation. When postlarvae or small shrimp are to be PCR tested, it is recommended to make a whole-body homogenate in order to then take an aliquot for RNA extraction and a subsequent PCR analysis for TSV genomic detection. Hepatopancreas, midgut or feces <u>should not</u> be used as a reliable tissue source to detect the presence of TSV.

Sampling and preservation of tissues for Shrimp MultiPathTM should be done in labelled vials and/or tubes that seal. The fixative should be 70% laboratory grade ethanol. Tissue samples should include pleopods, haemolymph of whole postlarvae supplying 2 to 5 mm² in size. Sampling equipment must be sterilized between samples.

Sampling numbers and health management plans should be established with your health expert who will take into account factors such as nauplii/postlarvae source, climate, farm size and location, company structure, market channels for sale of product, etc. Pooling shrimp samples for TSV testing to maximize value for money with PCR testing is routinely done.

Longer term solutions to disease caused by TSV include breeding for tolerance and resistance with biosecurity as a preventative strategy. Good sanitary and management farming practices may help to control the disease. These include:

- Improvement of maturation and hatchery sanitary molecular controls
- Frequent broodstock and postlarvae PCR-screening, with adequate broodstock management (specially females' prophylactic measurements)
- Use of TSV-negative postlarvae and good shrimp farm management including strict feeding rate control, reduction of organic matter in tanks and ponds, and appropriate stocking density
- Weekly-based surveillance for clinical signs of TSV causing-disease
- Early pathogen detection and risk mitigation through the use of Shrimp **Multi**Path™ is an important tool to mitigate disease.

It is worth noting that according to the WOAH (World Organisation of Animal Health), infected crustacean products can be treated at 70°C for 108 minutes to inactivate the pathogen.

Contact Genics at <u>info@genics.com</u> if you would like to discuss these options for your operation or visit <u>www.genics.com</u> for further details.

Learn how to dissect your shrimp for testing

Visit our **NEW Educational page** here to learn how to:

- Sterilize your equipment before sampling
- Selecting the correct ethanol for tissue preservation
- Identify and sample shrimp target organs for Shrimp **Multi**Path™ testing



Questions?

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Did you know?

Shrimp rarely harbour only one pathogen and farmers often don't know which ones they are. This is a significant economic risk for farmers. **Genics has solved this problem with** Shrimp MultiPathTM. It stacks up as the ultimate early warning system for farmers, **detecting up to 16 pathogens in a single test** that is unparalleled in today's industry for its sensitivity and accuracy.