



ICAR-CIBA - a nodal R&D agency working in brackishwater aquaculture for the past three decades with a vision of environmentally sustainable, economically viable and socially acceptable aquaculture technologies, system interventions, technology backstopping and policy inputs by the institute, contributing to economic benefits of the sector which has already recorded annual export revenue of ₹ 23,000 crores apart from domestic consumption.

Real time PCR Kit for diagnosis of Hepatopancreatic Microsporidiasis (HPM) of shrimps

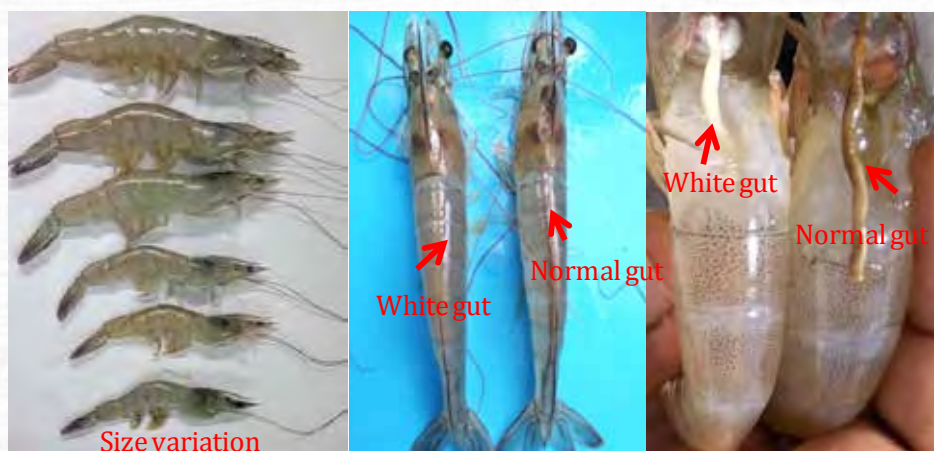
About the disease and the Pathogen

Hepatopancreatic microsporidiosis (HPM) caused by *Enterocytozoon hepatopenaei* (EHP) is a newly emerging disease of cultivated shrimp in Asia. *Enterocytozoon hepatopenaei* is a microsporidian belongs to the family Enterocytozoonidae. The spores are very small ($1.1 \times 0.7 \mu\text{m}$) and ultrastructure shows the presence of a polar filament of 5-6 coils. *Enterocytozoon hepatopenaei* was found to infect *Penaeus monodon*, *P. vannamei*. EHP has been reported from Thailand, Vietnam, Indonesia, Malaysia, China, Australia and India.

It infects only the tubular epithelial cells of the hepatopancreatic tissue of shrimp. Being the power house of the animal, infection in this digestive organ causes impairment of metabolism resulting in stunted growth. EHP infection can be transmitted horizontally through oral route (cannibalism, predation) and possibly by vertical transmission (trans-ovum). So far, no secondary hosts are known to be involved.

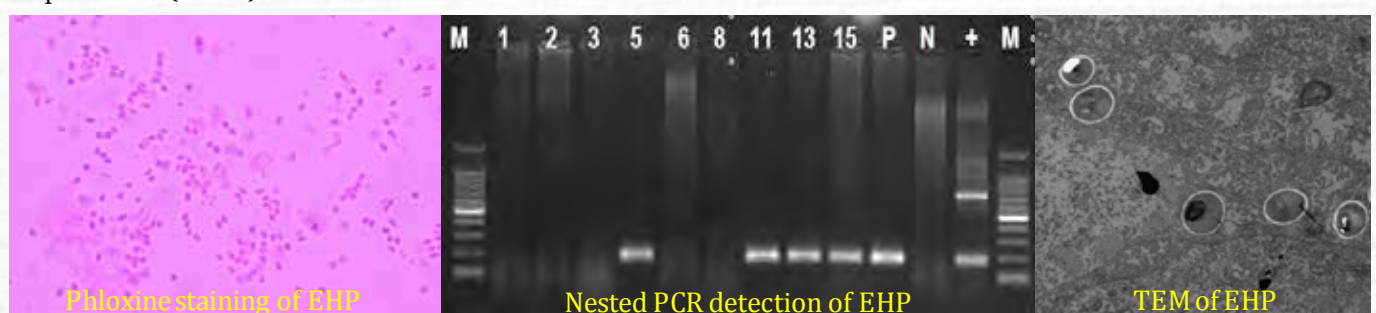
Clinical signs of Hepatopancreatic microsporidiasis

EHP infection in shrimp cannot be detected by visual inspection since no specifically distinctive gross signs are observed. However, suspected cases are often associated with growth retardation and white faeces syndrome.



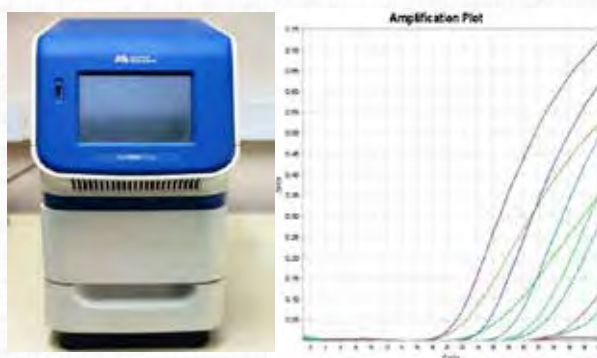
Detection of Hepatopancreatic microsporidiasis

EHP infection in shrimp can be detected by microscopically demonstrating spores (1.1 ± 0.2 by $0.6-0.7 \pm 0.1 \mu\text{m}$) in stained hepatopancreatic tissue smears, hepatopancreas tissue sections and faecal samples. Other diagnostic methods reported include histopathology, in situ hybridisation, polymerase chain reaction (PCR), loop-mediated isothermal amplification (LAMP) and real-time PCR



About the Kit

ICAR-CIBA is a lead research institution in the area of aquatic animal health in India and involved in development of fast, accurate and cost effective diagnostic kits for major shellfish and finfish pathogens in India. In this effort institute has developed a simple, cost-effective, rapid and sensitive diagnostic real time PCR assay kit. This assay is designed for the detection of EHP as low as 2 copies of viral DNA. This kit is cost effective, user friendly and does not require post PCR procedure. This assay also includes a house keeping gene that will ensure perfectness of PCR assay and nullifies any chances of false negative results due to failure in DNA extraction and presence of PCR inhibitors in the DNA.



Real time (qPCR) detection of EHP

Contents of the Kit:

- Kit package comes with either 50 or 100 reactions
- The kit includes reagents for DNA extraction and reagents for qPCR such as Real time master mix with Rox, Primer probe mix, negative and positive controls.
- Step by step protocol for performing the test



| Name of the kit | EHP real time PCR assay kit |
|--------------------|--|
| Intended use | Detection of EHP in shrimp tissue samples. |
| Test capacity | 50 or 100 reactions per kit |
| Sensitivity | 2 copies per reaction |
| Specificity | Specific to EHP and does not cross react with other shrimp pathogen. |
| Speed of diagnosis | 90 minutes (including DNA extraction) |
| Target users | Commercial aquaculture diagnostic and shrimp hatchery laboratories, Research organizations |
| Export potential | Yes |

“Brackishwater Aquaculture for Food, Employment and Prosperity”

ICAR-Central Institute of Brackishwater Aquaculture

(ISO 9001:2015 certified)

Indian Council of Agricultural Research,

75, Santhome High Road, MRC Nagar, Chennai 600 028 Tamil Nadu, India

Phone: +91 44 24618817, 24616948, 24610565 | Fax: +91 44 24610311

Web: www.ciba.res.in | Email: director.ciba@icar.gov.in, director@ciba.res.in

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