

Shrimple

Shrimp Virus Detection Kit

INTRODUCTION

Outbreaks of white spot syndrome (WSS), causing serious mortality to cultured shrimp, have been occurring in the East Asia since 1992. WSS has spread widely, and caused serious damage to shrimp farms in the world. Detection of white spot syndrome virus (WSSV) in shrimps is very important for management of shrimp farms.

EnBio Shrimp Virus Detection kit, named Shrimple, is a management tool based on immunoassay for shrimp aquaculture that can be used to assess the level of WSSV infection in larva and shrimp during breeding. Shrimple is a lateral flow, one step, fast and easy. The results are read visually without any instruments. The test system employs unique monoclonal antibodies to selectively identify WSSV in shrimps with a high degree of sensitivity.

PRINCIPLE OF THE TEST

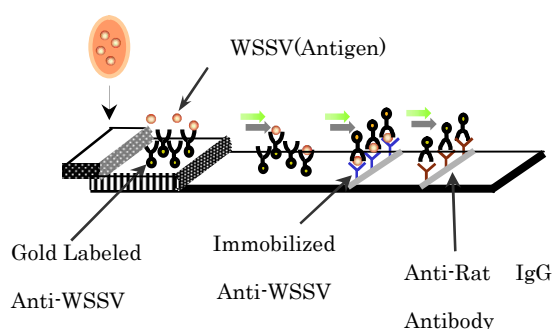
Shrimple is based on the principle of a sandwich immunoassay. The test device consists of a membrane strip and plastic case. A colored monoclonal rat anti-WSSV antibody colloid gold conjugate (antibody - colloid gold conjugate) pad is placed next to the sample pad region on the membrane strip. The membrane strip is also pre-coated with different monoclonal rat anti-WSSV antibody on the test (T) zone and anti-rat Immunoglobulin G (IgG) antibody on the control (C) zone.

In the absence of WSSV in the shrimp, the

antibody-colloid gold conjugate moves with the sample by capillary action along the membrane through the T zone until it reaches the C zone. At this point, antibody-colloid gold conjugate reacts with the pre-coated anti-rat IgG antibody to produce a visible pink colored band. When only one pink colored band appears on the C zone, it means negative.

When the WSSV is present in the shrimp, the antibody-colloid gold conjugate reacts with WSSV to produce conjugate-WSSV complex and moves until it reaches the T zone. At this point, the complex reacts with the pre-coated different monoclonal rat anti-WSSV antibody to produce a visible pink colored band. An excess free antibody-colloid gold conjugate passes through the T zone until the C zone. At this point, the excess antibody-colloid gold conjugate reacts with the pre-coated anti-rat IgG antibody to produce a visible pink colored band. When the pink colored band appears in both T zone and C zone, it means positive.

ILLUSTRATION OF MECHANIZUMU



Shrimple

Shrimp Virus Detection Kit

KIT CONTENTS

Each kit consists of following parts.

- Test Device
- Microtube with 0.6ml buffer liquid (20mM, TBS)
- Stick
- Dropper

SPECIMEN COLLECTION

Shrimps that look sick or unhealthy should be used. No special pre-treatment is necessary. The specimen collection may be stored up to 2 days at refrigerated condition (2-8 °C). If it is necessary to store the specimen more than 3 days, it should be stored under frozen condition (-20 °C or below)

Note: Do not use shrimps stored in alcohol as they adversely affect the test results.

TEST PROCEDURE

(Please refer the illustration of test in a last page.)

1. Count the number of dead shrimps with each culture pond everyday. When the number is more than usual or a lot of shrimps are floating on the surface of the water, pick up some of the shrimps that look sick or unhealthy.

Note: It is recommended that max 10 tests per 1 time be conducted with unhealthy or dead shrimps in the nursery over once a week even if the nursery has a well maintained.

2. Wash the shrimps should be tested with clean water and prepare the testing samples according to the section 3 and put them into a microtube. When the swimming legs are cut off at the bottom by hands, wash your hands well beforehand.

3. The number of testing sample required of one microtube is as follows:

Size of shrimp	Testing sample	Required number of shrimps
PL7	Whole body	20
PL10 or less	Whole body	10
PL20 or less	Whole body	5
0.05g or less	Whole body	2
0.1g or less	Whole body	1
0.5g or less	Whole body without head and carapace	1
2g or less	All legs	1
3g or less	All swimming legs	1
5g or less	Five swimming legs	1
10g or less	Three swimming legs	1
20g or less	Two swimming legs	1
Over 20g	One swimming leg	1

Note: Adjust sample volume to about 0.1g each 1 test

Shrimple

Shrimp Virus Detection Kit

4. Pulverize well the testing samples in the microtube by sticking them with the stick more than ten times. Then settle them for five minutes and take the supernatant with the dropper and put four droppings into the round hole of the Shrimple device.

Wait for 15 minutes and read the results. See followings for the interpretation of the results..

INTERPRETATION OF RESULTS

Negative: Only one pink colored band appears in the C zone. (In rare case, there is appearance of weak and non-pink colored band in the T zone, according to environmental influence. It means Negative result.)

Positive: In addition to one pink colored band in the C zone, a distinct pink colored band will also appear in the T zone.

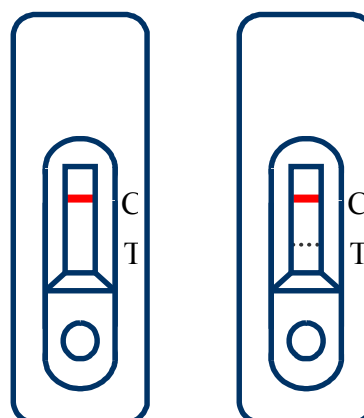
Invalid: No pink colored band appears in the C zone. An invalid result may be due to improper testing procedures or deterioration of the kit components. Repeat the assay sequence using a new device.

Note: Read the test result within 15 minutes as the band becomes indistinguishable with its color getting thinner with time going by

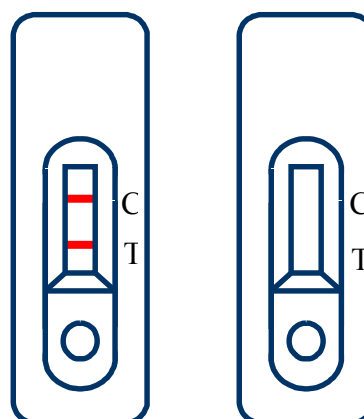
Note: When at least one tests shows positive, assume that many of the shrimps in the culture pond are infected. Such

positive result means the dangerous possibility that a mass death will occur in the following one week is high..

When up to ten tests show negative, you may assume that the shrimps in the nursery are not infected or that there will be less likelihood of mass death.



Negative



Positive

Invalid

Shrimple

Shrimp Virus Detection Kit

STORAGE AND STABILITY

The kit can be stored at room temperature in the sealed pouch to the date of expiration. The kit should be kept away from direct sunlight, moisture and heat. Expiration date is described on the product package.

PRECAUTION

1. For detecting WSSV in the larva and shrimp only.
2. The test device should not be reused.
3. The buffer liquid in the microtube should be used for virus testing purpose only. Avoid contact with skin or eyes.
4. Do not use test kit beyond the expiry date.
5. Discard the used kit in a safe place.

Important: Technical information contained herein is furnished without charge or obligations, and is given and accepted at recipient's sole risk.

EnBioTec Laboratories Co.,Ltd. is not responsible for any data and/or results with this kit because the conditions of use kit may vary and are beyond our control.

ILLUSTRATION OF TEST

