

MB581MPF32200 HiPurA[®] Pre- filled Plates For Shrimp DNA Purification
[For InstaNX[®] Mag32]

Kit Contents

Product Code	Reagents provided	MB581MPF32200
		10 NO
PF1L	Pre-filled Plate for Shrimp DNA extraction	20 nos
LA1096A	Magnetic Rod Tip for Insta NX [®] Mag32	40 nos
DS0015	Lysis Solution (AL)	260 ml
DS0013	Proteinase K	6.5 ml
DS0040	Elution Buffer (ET) [10 mM Tris-Cl, pH 8.5]	350 ml
DS1005A	Magnetic Beads	6.5 ml

Intended Use

Recommended for isolation of DNA from Shrimp sample.

Introduction

Shrimp is an important commodity in international trade accounting for 15% in terms of value of internationally traded seafood products. Aquaculture contributes to over 50% of global shrimp production. Shrimp production by aquaculture has been seriously impacted by diseases. The major constraints faced by shrimp aquaculture is the loss due to viral and bacterial diseases like white spot syndrome, yellow head disease, Taura syndrome, *Vibrio* spp., etc. HiMedia's HiPurA[®] Pre- filled Plates For Shrimp DNA Purification provides a fast and easy method for purification of total DNA for reliable applications in PCR technique etc. The DNA obtained is compatible with downstream applications such as restriction enzyme digestion, PCR and Southern blotting.

HiPurA[®] Pre- filled Plates For Shrimp DNA Purification

This kit simplifies isolation of DNA from shrimp tissue samples with magnetic based procedure. Shrimp tissue (spliced and digested) is subjected to lysis by Proteinase K in a chaotropic salt solution. Following lysis, is the adsorption of nucleic acid to the magnetic beads, removal of residual contaminants and elution of pure nucleic acid. The magnetic beads have a high binding capacity and high-quality DNA is obtained from shrimp samples.

Elution

The yield of genomic DNA depends on the sample type and the number of cells in the sample. A single elution with 100 µL of Elution Buffer (ET) will provide sufficient DNA to carry out multiple amplification reaction. Elution with volume less than 100 µL will increase the final DNA concentration, but will reduce the overall DNA yield.

Storage

HiPurA® Pre- filled Plates For Shrimp DNA Purification can be stored at room temperature (15-25°C) for up to 1 year without showing any reduction in performance.

Materials needed but not provided

- Tabletop Microcentrifuge (with rotor for 2.0 ml tubes)
- Ethanol (96 - 100%)
- Ethanol (70%)
- Insta NX® Mag32 (Product Code: LA1096)
- Vortex
- Polypropylene sealing film (Product Code: PR21)
- Collection Tube, 2.0ml (Product Code: PW1139)
- Sterile plastic sachets and pestle (for tissue homogenization)

General Preparation Instructions

1. Thoroughly mix reagents

Examine the reagents for precipitation. If any kit reagent forms a precipitate (other than enzymes), warm at 55-65°C until the precipitate dissolves and allow cooling to room temperature (15-25°C) before use.

2. Ensure that clean & dry Nuclease-free tubes and tips are used for the procedure.

Procedure

1. Prepare tissue

Weigh 100mg of fresh or frozen tissue and mince quickly. If frozen tissue is used, allow it to thaw slightly before slicing but keep on ice in order to protect degradation. Cut the tissue into small pieces as it enables more efficient lysis.

NOTE: Tissue can be harvested by aliquoting in 2.0 ml collection tubes and flash freezing in liquid nitrogen; these can be stored at -70°C for several months before preparing DNA.

2. Digest tissue

In a sterile plastic sachet, add 800 µL of Lysis Solution (AL) (DS0015) and 20 µL of Proteinase K solution to the tissue. Homogenize the tissue by grinding it with a pestle. Incubate the sample at room temperature (15- 25°C) for 10 minutes.

3. Transfer the homogenized mixture to a 2 ml micro-centrifuge tube and centrifuge at 5000rpm for 5 minutes.

4. Transfer 150 µL of supernatant to fresh tube and add 450 µL of 100% chilled ethanol. Mix well by vortexing and centrifuge at 10,000 rpm for 5 minutes at 4°C.

5. Wash

Discard the supernatant and re-suspend the pellet with 400 µL of 70% ethanol and centrifuge at 13,000 rpm for 5 minutes at 4°C.

6. Discard the supernatant and dry the pellet. Resuspend the DNA pellet in 100 µl of Elution Buffer (ET) (DS0040). **This will be pre-processed sample.**

Set up processing plates as follows:

1. Switch on the UV light for 10 minutes prior to use.
2. Select “**MB581Shrimp**” program.
3. Place the magnetic rods tip by sliding onto the machine.

NOTE: After placing the rods ensure that the rods are properly fixed on their place.

4. Remove the seal from the Pre-filled Plate for Shrimp DNA extraction (**PF1L**).

NOTE: Take care while peeling off the seal. Hold the plate firmly by one hand and then peel off the seal by holding it in your other hand without shaking the plate.

5. Add **100µl pre- processed sample** into **1st well** and **7th well** of the Pre-filled Plate for Shrimp DNA extraction (**PF1L**).
6. **Add 20µl Magnetic Beads (DS1005A) in the 1st and the 7th column of the Pre-filled Plate for Shrimp DNA extraction(PF1L) for Insta NX[®] Mag32.**
7. Slide the tray in outward direction and after adding the above solutions place the plates on the tray of the machine.

NOTE: 16 samples can be processed in a single Pre-filled Plate for Shrimp DNA extraction(PF1L).

8. Slide the tray of the machine back to its position and close the door of Insta NX[®] Mag32 machine.
9. Click on the **RUN** option on the home screen.
10. After the run is complete, slide the tray of the machine in outward direction. Remove the 96 Deep Well Plate from the position. Slide the tray back to its position and discard the Magnetic rod’s tip for Insta NX[®] Mag32 (LA1096A). Dispense the eluted nucleic acid from column 6 and column 12 to a new Collection Tube, Polypropylene (2.0 ml) (PW1139) (not provided). The eluate contains pure nucleic acid.

NOTE: A small amount of magnetic beads may be observed in the final eluate at the bottom of the plate. Avoid transferring these magnetic beads to your PCR reaction mixture.

Storage of the eluate with purified nucleic acid: The eluate contains pure genomic DNA. For short-term storage (24-48 hrs) of the DNA, 2-8°C is recommended. For long-term storage, -20°C or lower temperature (-80°C) is recommended. Avoid repeated freezing and thawing of the sample which may cause denaturing of DNA. The Elution Buffer will help to stabilize the DNA at these temperatures.

Limitations

The yield of nucleic acid depends upon the type and the volume of starting material used.

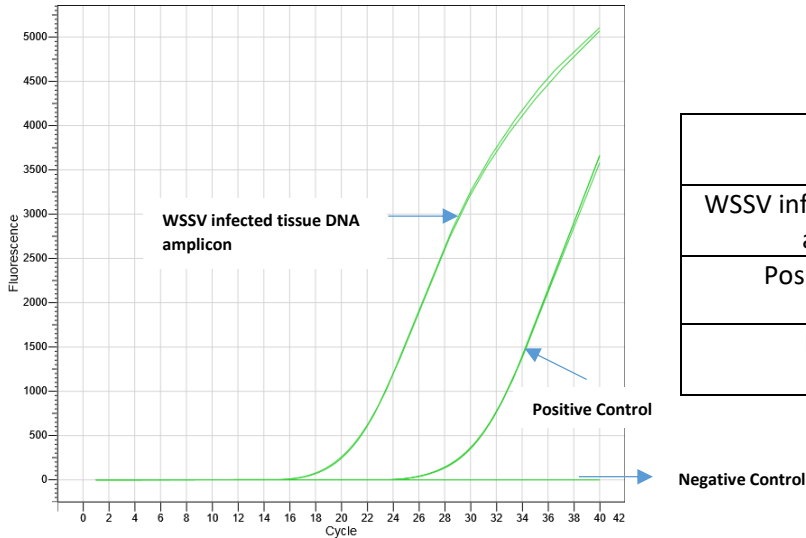
Performance and Evaluation

The yield and efficiency of purification is determined by performing Real- Time PCR.

Quality Control

Type of Sample	DNA Purity
WSSV infected tissue	1.6-1.9

Amplification Data



Sample	Ct value	Fluorescence Value
WSSV infected tissue DNA amplicon	21.46	5000
Positive Control	30.74	3500
Negative Control	N/A	N/A

Image representing probe based real-time amplification data of WSSV DNA extracted using MB581MPF32200- HiPurA® Pre-filled Plates for Shrimp DNA Purification, with Ct values (provided in table). The results completely depend upon sample types.

Each lot of HiMedia’s HiPurA® Pre- filled Plates For Shrimp DNA Purification is tested against predetermined specifications to ensure consistent product quality.

Warning

Not for Medicinal Use. Read the procedure carefully before beginning the protocol. Wear protective gloves/protective clothing/eye protection/face protection. Follow good clinical laboratory practices while handling clinical samples. Standard precautions should be followed as per established guidelines. Safety guidelines may be referred in safety data sheets of the product.

Please refer disclaimer Overleaf.

Safety Information

The HiPurA® Pre- filled Plates for Shrimp DNA Purification is for laboratory use only, not for drug, household or other uses. Take appropriate laboratory safety measures and wear gloves when handling. Not compatible with disinfecting agents containing bleach. Please refer the Safety Data Sheet (SDS) for information regarding hazards and safe handling practices.

Disposal

User must ensure safe disposal by autoclaving and/ or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques.

Technical Assistance

At HiMedia, we pride ourselves on the quality and availability of our technical support. For any kind of technical assistance mail to mb@himedialabs.com.



Storage temperature



Do not use if package is damaged



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