

MB505PC16 HiPurA® Pre- filled Cartridges for Bacterial DNA Purification

Kit Contents

Product Code	Reagents provided	MB505PC16
		48 PR
PF16U	Pre-filled Cartridges for Bacterial DNA purification	48 no
LA1118B	Magnetic Rod Tip	12 no
DS1005A	Magnetic Beads	1 ml
DS0003	RNase A Solution (20mg/ml)	1 ml
DS0013	Proteinase K	1 ml
DS0040	Elution Buffer (ET) [10mM Tris-Cl, pH8.5]	3.5 ml
DS0015	Lysis Solution (AL)	9 ml

Intended Use

Recommended for isolation of DNA from bacterial cultures.

Introduction

HiPurA® Pre- filled Cartridges for Bacterial DNA Purification provides the fastest and easiest way to purify DNA for reliable use in amplification technologies. HiPurA® Pre- filled Cartridges for Bacterial DNA Purification can be used for isolation of genomic DNA from a wide variety of samples, but the performance may vary depending on the sample type.

HiPurA® Pre- filled Cartridges for Bacterial DNA Purification Kit

This kit carries out efficient extraction of genomic DNA from wide range of samples. Sample is first lysed under the highly denaturing conditions provided by Lysis Solution to inactivate DNases and to ensure isolation of intact genomic DNA. The DNA purification procedure comprises of three steps viz. adsorption of DNA to the magnetic beads, removal of residual contaminants and elution of pure genomic DNA. The magnetic beads have a high binding capacity and high-quality genomic DNA is obtained from various species. The DNA obtained is compatible with downstream applications such as restriction enzyme digestion, PCR and Southern blotting.

Elution

The yield of DNA depends on the sample type and the number of cells in the sample. A single elution with Elution Solution will provide sufficient DNA to carry out multiple amplification reactions.

Storage

HiPurA® Pre- filled Cartridges for Bacterial DNA Purification can be stored at room temperature (15-25°C) for up to 2 years without showing any reduction in performance. We advise a certain storage temperature for the reagents listed below:

- **On receipt store Proteinase K (DS0013): at -20°C.**
- **On receipt store RNase A (DS0003): at 2-8°C.**
- **On receipt store Magnetic Beads (DS1005A): at 2-8°C.**

Materials needed but not provided

- 55°C water bath or heating block (For Lysate Preparation)
- Tabletop Microcentrifuge (with rotor for 2.0 ml tubes) TabSpin® 012 (LA1090)
- Insta NX® Mag16 (LA1118)
- Cartridge Holder (LA1118CH)
- Vortex
- Polypropylene sealing film (Product Code: PR21)
- Centrifuge tubes, 2.0ml (Micro Centrifuge Tube-C- PW146)
- HiPer® Lock Microcentrifuge Tube, 2.0ml (Product Code: MBLA017)
- 1X PBS (ML116)
- Insta NX® Mag32 (LA1096)

General Preparation Instructions

1. Preheat a water bath or heating block to 55°C.
(For Lysate preparation)

2. **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.

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

4. Vortex magnetic beads before use.

RNase A enzyme treatment

RNase A is a type of RNase that is commonly used in research. RNase A (e.g., bovine pancreatic ribonuclease A) is one of the sturdiest enzymes in common laboratory usage. It cleaves 3' end of unpaired C and U residues.

Unit Definition for RNase A

One unit of the enzyme causes an increase in absorbance of 1.0 at 260 nm when yeast RNA is hydrolyzed at 37°C and pH 5.0. Fifty units are approximately equivalent to 1 Kunitz unit. It is completely free of DNases and proteases. The specific activity is 90 U/mg.

The product as supplied is stable at room temperature (15-25°C).

Specimen Handling and Collection

Collect overnight culture from sterile flask with the help of micropipette. Store the remaining culture at 2-8°C for short term use.

Type of Specimens

Bacterial Culture

Procedure

1. Harvesting of cells

Pellet 1 ml of an overnight bacterial broth culture in provided 2 ml capped collection tube (not provided) by centrifuging for 2 minutes at 13,000 rpm at room temperature (15-25°C). Remove the culture medium and discard.

NOTE: If bacteria are grown in rich media such as Terrific broth, it is necessary to reduce the volume of the starting material of the overnight broth culture to 0.5 ml in order to avoid overloading.

OR

2. Harvesting of colonies

Take small size 5-6 colonies or 2-3 medium to large colonies from culture plate in 1 ml of 1X PBS (ML116) (not Provided) to make homogeneous suspension. Pellet down the suspension by centrifuging for 2 minutes at 13,000 rpm at room temperature (15-25°C) and discard the supernatant without disturbing the pellet.

3. Resuspend cells

Resuspend the pellet thoroughly in 180 µl of Lysis Solution (AL) (DS0015).

4. Prepare for cell lysis

Add 20 µl of the Proteinase K (DS0013) solution to the sample. Mix and incubate for 30 minutes at 55°C. **This will be your pre- processed sample.**

Optional RNase A treatment

If RNA-free genomic DNA is required, add 20 µl of RNase A Solution (DS0003), mix and incubate for 5 minutes at room temperature (15-25°C)

Set up processing Cartridges as follows:

1. Switch on the UV light for 10 minutes prior to use.
2. Open the door of Insta NX® Mag16 machine.
3. Select “**MB505M**” program.
4. Remove the seal from the Pre-filled Cartridges for Bacterial DNA Purification (PF16U). Place the Pre-filled Cartridges for Bacterial DNA Purification (PF16U) into the Cartridge Holder (LA1118CH) (not provided).

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

5. Add 50µl of Elution Buffer (ET) [10mM Tris-Cl, pH8.5] (DS0040) into the 6th well of the Pre-filled Cartridges for Bacterial DNA purification (PF16U).
6. Add 200µl of pre- processed sample and 20µl of Magnetic Beads (DS1005A) in the 1st well of the Pre-filled Cartridges for Bacterial DNA purification (PF16U).

7. Place the Cartridge Holder (LA1118CH) along with cartridges on the platform. Place the Magnetic Rod Tip (LA1118B) by sliding onto the machine.

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

8. Close the door of Insta NX® Mag16 machine.
9. Click on the **RUN** option on the home screen.
10. After the run is complete, remove Pre-filled Cartridges for Bacterial DNA Purification (PF16U) from the position. Discard the Magnetic Rod Tip (LA1118B). Dispense the eluted DNA from well 6 to a new HiPer® Lock Microcentrifuge Tube, 2.0ml (MBLA017) (not provided). The eluate contains pure DNA.

NOTE: If small amount of magnetic beads are observed in the final eluate then keep the cartridges along with cartridge holder on Magnetic pad (not provided) for 4-5 minute and collect supernatant carefully without disturbing beads pellet in new collection tube.

OR

Take out eluate in new collection tube and centrifuge at higher speed for around 1 min to pellet down the traces of Magnetic beads present in the eluate.

***NOTE: If you process less than 4 samples at a time please order LA1118B- Magnetic Rod Tip (Pack size- LA1118B-4no/ LA1118B-40no).**

Storage of the eluate with purified DNA: The recommended storage temperature for the eluted DNA is -80°C. Avoid repeated freezing and thawing of the sample which may cause denaturing of DNA.

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.

Limitations

The yield of DNA 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

Each lot of HiMedia's HiPurA® Pre- filled Cartridges for Bacterial DNA Purification is tested against predetermined specifications to ensure consistent product quality.

Safety Information

The HiPurA® Pre- filled for Bacterial 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.

Please refer disclaimer Overleaf.










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 off 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.

Symbols

	Manufacturer		Do not use if package is damaged
	Catalogue number		Temperature limit
	Date of manufacture (YYYY-MM)		Consult instructions for use
	Use-by date (YYYY-MM)		Contains biological material of animal origin
	Batch code		

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Disclaimer :

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HiMedia Laboratories Pvt. Ltd. Reg.office : Plot No. C-40, Road No. 21Y, MIDC, Wagle Industrial Estate, Thane, (West) 400604, Maharashtra, INDIA. Customer Care No.: 00-91-22-6116 9797 Tel: 00-91-22-6147 1919, 6903 4800 Email: techhelp@himedialabs.com Website: www.himedialabs.com