

MB571PC16 **HiPurA® SuperPlant Pre- filled Cartridges for Plant DNA Extraction**

Kit Contents

Product Code	Reagents provided	MB571PC16
		48 PR
PF16X	SuperPlant Pre-filled Cartridges for Plant DNA Extraction	48 no
LA1118B	Magnetic Rod Tip	24 no
DS0200A	SuperPlant Extraction Buffer A	44 ml
DS0040	Elution Buffer (ET) [10 mM Tris-Cl, pH 8.5]	7 ml
DS0003	RNase A Solution (20mg/ml)	1 ml
DS1005A	Magnetic Beads	1 ml
DS0070	Additive-II	4.5 ml
DS0071	Additive-III	0.9 g

Intended Use

Recommended for isolation of DNA from plant samples.

Introduction

HiPurA® SuperPlant Pre- filled Cartridges for Plant DNA Extraction for Insta NX® Mag16 provides the fastest and easiest way to purify DNA for reliable use in amplification technologies. HiPurA® SuperPlant Pre- filled Cartridges for Plant DNA Extraction can be used for isolation of DNA from various samples, but the performance may vary depending on the sample type. The DNA purification procedure comprises of three steps viz. adsorption of DNA to the magnetic beads, removal of residual contaminants and elution of pure DNA. The magnetic beads have a high binding capacity and high-quality DNA is obtained from sample. The purified DNA can directly be used for PCR analysis and other downstream applications.

HiPurA® SuperPlant Pre- filled Cartridges for Plant DNA Extraction

This kit carries out efficient extraction of DNA from plant samples. The procedure is optimized for a maximum of 200 mg of wet-weight of the starting material. The sample is cut and ground in liquid nitrogen or without liquid nitrogen along with SuperPlant Extraction Buffer A. The flow-through fraction is then mixed with a solution that enhances the binding of DNA to the magnetic particles. The magnetic particles are then carried forward to the washing step to remove trace contaminants. High quality DNA is eluted in the Elution Buffer (ET) provided in the pre-filled Cartridges.

Elution

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

Storage

HiPurA® SuperPlant Pre- filled Cartridges for Plant DNA Extraction 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 RNase A (DS0003): at 2-8°C.**
- **On receipt store Magnetic Beads (DS1005A): at 2-8°C.**

Materials needed but not provided

- Small mortar and pestle
- Liquid nitrogen (Optional)
- Tabletop Microcentrifuge (with rotor for 2.0 ml tubes) capable of upto ≈13,000 rpm [TabSpin® 012 (LA1090)]
- 65°C water bath or heating block
- Insta NX® Mag16 (Product Code: LA1096)
- Insta NX® Mag32 (LA1096)
- Vortex
- HiPer® Lock Microcentrifuge Tube, 2.0ml (Product Code: MBLA017)
- Chloroform: Isoamylalcohol (24:1) (Product Code: MB115)
- Cartridge Holder (LA1118CH)

General Preparation Instructions

1. Grinding of the plant material can be done using mortar and pestle. Midrib should be removed from the material before grinding, as midrib is a major source of carbohydrate contamination.
2. **SuperPlant Extraction Buffer:** Immediately prior to use, add 90 µl of Additive-II and 18 mg of Additive-III in 900 µl of SuperPlant Extraction Buffer. Preheat the solution to 65°C.

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 260nm 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

Sample Preparation with liquid Nitrogen

Finely cut the leaf material before grinding. Weigh 200 mg of the finely cut plant material and grind properly using a mortar and pestle in liquid nitrogen to a fine powder. Allow the liquid nitrogen to evaporate. **DO NOT ALLOW THE SAMPLE TO THAW** (keep samples on ice if needed). **Proceed immediately to the DNA isolation protocol (Pre- processing of samples).**

OR

Sample Preparation without liquid Nitrogen

Finely cut the leaf material before grinding. Weigh 200 mg of the finely cut plant material and grind properly using a mortar and pestle 900 µl of SuperPlant Extraction Buffer A (DS0200A) (preheated to 65°C) (Refer General Preparation Instructions). **Proceed immediately to the DNA isolation protocol with step 2.**

Pre- processing of samples

NOTE: Ensure that Additive-II and Additive-III are added to SuperPlant Extraction Buffer A (DS0200A) as mentioned in General Preparation Instructions.

1. To 200mg of the ground material (with liquid nitrogen) add 900µl of SuperPlant Extraction Buffer A (DS0200A) (preheated to 65°C) (Refer General Preparation Instructions) and transfer the sample to a capped 2.0 ml collection tube (not provided). Mix by vortexing.
2. Transfer the mixture to a capped 2.0 ml collection tube and incubate the samples for 60-90 minutes with occasional inversion at 65°C.
3. Add 1ml of Chloroform: Isoamylalcohol (24:1) (not provided) and mix gently by inversion for 5 minutes.
4. Centrifuge the samples at 13,000 x g [≈14,000 rpm] for 10 minutes at room temperature (15-25°C).
5. Transfer the top aqueous layer (containing DNA) into a fresh 2.0ml collection tube (not provided) and add 20µl of RNase A Solution (20 mg/ml) (DS0003). Incubate for 5 minutes at room temperature (15-25°C). **This will be your pre- processed sample.**

Set up processing Cartridges as follows:

1. Switch on the UV light for 10 minutes prior to use.
2. Select “**MB57116**” program. Open the door of the Insta NX® Mag16 machine.
3. Remove the seal from the SuperPlant Pre-filled Cartridges for Plant DNA Extraction (PF16X).

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

4. Add **100µl of Elution Buffer (ET)** [10mM Tris-Cl, pH8.5] (DS0040) into the **6th well of the SuperPlant Pre-filled Cartridges for Plant DNA Extraction (PF16X).**
5. Add approximately **450µl pre- processed sample** in the **1st & 2nd well of the Super Plant Pre-filled Cartridges for Plant DNA Extraction (PF16X).**
6. **Add 10µl Magnetic Beads (DS1005A) in the 1st & 2nd well of the SuperPlant Pre-filled Cartridges for Plant DNA Extraction (PF16X).**
7. Place the Magnetic Rods Tip (LA1118B) onto the machine.

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

8. Close the door and Click on the **RUN** option on the home screen.

9. After the run is complete, remove Cartridge Holder (LA1118CH) & cartridge 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 (Product Code: 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.

Storage of the eluate with purified DNA: The eluate contains pure DNA. For short-term storage (24-48 hrs.) of the DNA, 2-8°C is recommended. For long-term storage, recommended to be stored at -20°C or lower temperature (-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® SuperPlant Pre- filled Cartridges for Plant DNA Extraction is tested against predetermined specifications to ensure consistent product quality.

Safety Information

The HiPurA® SuperPlant Pre- filled Cartridges for Plant DNA Extraction 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.

Please refer disclaimer Overleaf.

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