MAGBIO

"WE MAKE NGS BETTER"

HighPrep DTR - DX

Dye Terminator Removal/Cleanup

Catalog Nos. DT-70001E, DT-70005E, DT-70050E, DT-70250E, DT-70500E
Manual Revision 0
WI-72-61

- Magnetic beads based chemistry
- No centrifugation or filtration

Instructions For Use

Contents

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CE IVD MHRA

For *in vitro* diagnostic procedures.
Information in this document is subject to change without notice.

Product Description

HighPrep DTR - DX uses a simple 3 steps procedure: Bind-Wash-Elute. Sanger sequence reaction clean-up process involves a selective binding of DNA to the HighPrep DTR - DX particles, followed by the removal of unincorporated dye terminators, nucleotides, salts, primers, primer dimers, and other contaminants. DNA amplicons and fragments are selectively bound to the magnetic beads' particles; and highly purified DNA is eluted with low salt elution buffer or water which can be used directly for downstream applications. HighPrep DTR - DX is designed for both manual and fully automated purification of dye terminators prior to sequencing.

Process

HighPrep DTR - DX uses a magnetic bead-based chemistry to remove unincorporated terminators and salts from Sanger sequencing reactions. The unincorporated terminators are often referred to as dye blobs, and they interfere with sequence analysis. The kit is intended for robust and efficient amplicon cleanup to produce pure DNA ready for sanger sequencing.

Product Specifications

Catalog Number	Description	Number of Reactions	Storage Conditions
DT-70001E	HighPrep DTR - DX (1 mL)	100	2-8°C DO NOT FREEZE
DT-70005E	HighPrep DTR - DX (5 mL)	500	
DT-70050E	HighPrep DTR - DX (50 mL)	5,000	
DT-70250E	HighPrep DTR - DX (250 mL)	25,000	
DT-70500E	HighPrep DTR - DX (500 mL)	50,000	

Number of reactions is based on a 10 µL reaction volume.

Storage and Preparation

- Store at 2-8°C. **DO NOT FREEZE.**
- Keep at room temperature for 30 minutes prior to use.
- Thoroughly shake the HighPrep DTR - DX to resuspend the beads before use.

Materials Supplied in the Kit

- HighPrep DTR - DX

Equipment and Reagents to Be Supplied by the User:

- 85% Ethanol (Prepare from absolute Ethanol. Do not use denatured alcohol)
- Polypropylene reservoirs
- Elution buffer (10 mM Tris pH 8.5, TE Buffer, 0.1mM EDTA, or diH_2O)
- Magnetic separation device compatible with 96 or 384 well PCR plate (see page 4)
- 96-well PCR plate or 384-well PCR plate
- Multichannel pipette

HighPrep DTR - DX: Dye terminator removal/cleanup using a 96-well plate

⚠ Bring the **HighPrep DTR - DX** to room temperature for at least 30 min before use.

1. Thoroughly shake the **HighPrep DTR - DX** to fully resuspend the magnetic beads.

2. Add 10 µL of **HighPrep DTR - DX** to each sample.
   Use 10 µL of **HighPrep DTR - DX** regardless of the volume of the sequencing reaction.

3. Add freshly prepared 85% Ethanol volume according to the table below:

Reaction Volume (µL)	85% Ethanol (µL)*
5	30
10	40
15	50
20	60

* Do not use denatured alcohol. Prepare fresh from absolute Ethanol.

4. Mix well the **HighPrep DTR - DX** and sample by pipetting up and down 7-10 times.

5. Place the sample plate on the 96 magnetic separation device for 4-5 minutes or until the solution clears. Beads will pull to the side of the well.

6. With the plate still on the magnet, remove and discard the supernatant by pipetting.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

7. Keep the sample plate on the magnet, add 100 µL of 85% Ethanol to each well, and wait 1-2 minutes or until the magnetic beads are fully resettled. Mixing is not necessary.

8. With the sample plate still on the magnet, remove and discard the supernatant by pipetting.

9. Repeat steps 7-8 for a total of two 85% Ethanol washes.

10. Dry the beads by incubating for 10 minutes at room temperature with the plate still on the magnetic separation device.

    ⚠ *It is critical to completely remove all liquid from each well since it contains traces of excess fluorescent dye and other contaminants.*

11. Remove the sample plate from the magnet. Add 40 µL of appropriate elution buffer (10 mM Tris pH 8.5, TE Buffer, 0.1mM EDTA, or diH_2O) to each well and pipette up and down 20 times to mix.

12. Incubate at room temperature for 5 minutes.

13. Place the sample plate back on the magnetic separation device and wait 5-7 minutes or until the magnetic beads clear from the solution.

14. Transfer the eluate (cleared supernatant) to a new plate to be loaded on a sequencer or store at -20°C for later use.

HighPrep DTR - DX: Dye terminator removal/cleanup using a 384-well plate

⚠ Bring the **HighPrep DTR - DX** to room temperature for at least 30 min before use.

1. Thoroughly shake the **HighPrep DTR - DX** to fully resuspend the magnetic beads.

2. Add 5 µL of **HighPrep DTR - DX** to each sample.
   Use 5 µL of **HighPrep DTR - DX** regardless of the volume of the sequencing reaction.

3. Add freshly prepared 85% Ethanol volume according to the table below:

Reaction Volume (µL)	85% Ethanol (µL)*
5	14.3
10	21.4
15	28.6

* Do not use denatured alcohol. Prepare fresh from absolute Ethanol.

4. Mix well the **HighPrep DTR - DX** and sample by pipetting up and down 7-10 times.

5. Place the sample plate on the 384 magnetic separation device for 3-4 minutes or until the solution clears. Beads will pull to the side of the well.

6. With the plate still on the magnet, remove and discard the supernatant by pipetting.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

7. Keep the sample plate on the magnet, add 30 µL of 85% Ethanol to each well, and wait 1-2 minutes or until the magnetic beads are fully resettled. Mixing is not necessary.

8. With the plate still on the magnet, remove and discard the Ethanol by pipetting.

9. Repeat steps 7-8 for a total of two 85% Ethanol washes.

10. Dry the beads by incubating for 10 minutes at room temperature with the plate still on the magnetic separation device.

    ⚠ *It is critical to completely remove all liquid from each well since it contains traces of excess fluorescent dye and other contaminants.*

11. Remove the sample plate from the magnet. Add 15-20 µL of appropriate elution buffer (10 mM Tris pH 8.5, TE Buffer, 0.1mM EDTA, or diH_2O) to each well and pipette up and down 20 times to mix.

12. Incubate at room temperature for 5 minutes.

13. Place the sample plate back on the magnetic separation device and wait 5-7 minutes or until the magnetic beads clear from solution.

14. Transfer the eluate (cleared supernatant) to a new plate to be loaded on a sequencer or store at -20°C for later use.

Ordering

HighPrep DTR - DX

Catalog No.	Product
DT-70005E	HIghPrep DTR - DX (5 mL)
DT-70050E	HIghPrep DTR - DX (50 mL)
DT-70250E	HIghPrep DTR - DX (250 mL)
DT-70500E	HIghPrep DTR - DX (500 mL)

Related Products

gDNA Isolation Kit

Catalog No.	Product	Description	Preps
HPBTS-D96E	HighPrep Blood & Tissue DNA Kit - DX (96 Preps)	Genomic DNA isolation from 20-250 µL of blood, lysate of tissues, mouse tails, cultured cells, or buccal swabs.	96
HPBTS-D96X4E	HighPrep Blood & Tissue DNA Kit - DX (384 Preps)	Genomic DNA isolation from 20-250 µL of blood, lysate of tissues, mouse tails, cultured cells, or buccal swabs.	384

Magnetic Separation Devices

Catalog No.	Description
MYMAG-96	Handheld Magnetic Separation Device (96 well microplate format)
MYMAG-96X	Magnetic Separation Device (96 well ring format)
MBMS-12	MagStrip Magnet Stand (1.5 mL x 12)
MBMS-31550	15 mL and 50 mL Magnetic Stand Combo (3 x 15 mL and 3 x 50 mL)

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