PERFORMA® DTR V3 96-Well Short Plates

Product	Catalog #	Purifications
Performa DTR V3 96-Well Short Plates (10 Plates)	47938	960
Performa DTR V3 96-Well Short Plates (50 Plates)	80808	4800

Description

Performa DTR (**D**ye **T**erminator **R**emoval) V3 96-Well Short Plates are gel filtration plates that consist of 440-µl volume columns in a standardized array. This plate provides optimal performance for removal of BigDye® v3.0 and v3.1. along with other dye terminators, dNTPs, salts and other low molecular weight materials from sequencing reactions. These columns also remove DNA primers and fragments up to 15 bases, buffers, and nucleotides labeled with biotin, isotopes and other assorted markers.

The columns are pre-packed with a fully hydrated matrix to afford optimal handling and performance characteristics. To minimize the potential for interference with sequencing applications, no preservatives, salts or buffers are used in the preparation of these columns. Both ends of the Performa 96-Well Short Plate are sealed to prevent drying.

The sample can be spun directly into the ABI PRISM® MicroAmp® Optical 96 Well Reaction Plate or equivalent (96-Well Capillary Plates), thereby saving a transfer step.

Component	47938	80808
Performa DTR V3 96-Well Short Plates	10 plates (10 x PN 4050203)	50 plates (50 x PN 4050203)

Equipment and Materials Required

1. Variable speed centrifuge (benchtop or floor model)
2. Rotor and microplate carriers for above.
3. 96-well receiver[1] and waste[2] plates
4. 96-Well Plate Lids (Edge BioSystems Cat. No. 33100)

Storage Condition

Store at +4°C. Do not freeze.

Quality Control

Field-tested for sequence quality and sequencing accuracy on a capillary sequencer.

Recommended Protocol for 5 µl–15 µl Sequencing Reaction Volumes

1. **Bring reaction volume to at least 10 µl with distilled water before adding to the V3 96-Well Short Plate.**
2. **Remove the bottom and top adhesive tapes from a PERFORMA V3 96-Well Short Plate. Cover with lid.**
   - Note: Remove the bottom adhesive tape first.
   - Ensure that the plate remains horizontal to avoid losing any gel.
3. **Stack the V3 96-Well Short Plate on top of a 96-well waste plate[2]. Place assembly on a cushioned centrifuge carrier.**
4. **Centrifuge for 3 minutes at 850 *x g*.[3] Discard eluate.**
   - See "Additional Notes" for determination of RPM from RCF or visit our website at www.edgebio.com and click on Technical Support.
5. **Transfer the reaction samples in a volume of 10-15 µl to the center of each well in the 96-Well Short Plate. Pipet slowly. Do not touch the sides of the wells. Cover with lid.**
6. **Stack the V3 96-Well Short Plate on top of a 96-well receiver plate[1]. Place the assembly on a cushioned centrifuge carrier.**
7. **Centrifuge for 5 minutes at 850 *x g*. Retain eluate.**
   - The eluate contains purified sample ready for loading on sequencers.
   - Note: Consult the instrument manufacturer's recommendation for sample handling.

Additional Notes

1. The following receiver plates are recommended: Edge Biosystems 96-Well Capillary Plate (Cat. No 13506), ABI MicroAmp® Optical 96 Well Reaction Plate, Nunc Polypropylene V-bottom plate (part # 442587) and Costar Polystyrene V-bottom plate (part # 3897) .

2. The following waste plate is recommended: Costar Flat Bottom plate (part # 9017).
   - The waste and receiver plates should be of the same height to allow the use of identical centrifuge setting for both steps 4 and 7.

3. Conversion of RCF to RPM Calculation:

   An accurate determination of the centrifugation speed is very important. The relative centrifugal force (RCF) specified in the protocol is converted to revolutions per minute (RPM) using the following formula:

$$RCF = 1.12\, r \left(\frac{RPM}{1000} \right)^2$$

   The radius, r, is equal to the distance in millimeters between the axis of rotation and the bottom of the gel bed when the plate is placed in the plate carrier in the centrifuge bucket.

   After measuring the radius for the specific centrifuge and accessories to be used, the proper RPM setting is calculated as follows:

$$RPM = 1000 \sqrt{\frac{RCF}{1.12\, r}}$$

   To achieve RCF = 850 *x g*:

$$\boldsymbol{RPM} = 27{,}549 \sqrt{\frac{1}{r}}$$