EXCELAPURE 96-Well UF PCR Purification Plates

Product	Catalog #	Purifications
ExcelaPure 96-Well UF PCR Purification Plates (10 Plates)	36181	960
ExcelaPure 96-Well UF PCR Purification Plates (50 Plates)	95674	4800

Description

ExcelaPure 96-Well UF PCR Purification Plates provide a simple, automatable, high-throughput method for purifying PCR reactions in 50-300µl volumes using ultrafiltration. ExcelaPure 96-Well UF Plates consist of an optimized ultrafiltration membrane for high recovery of your PCR products. Excess primers, primer dimers, dNTPs and salts are filtered to waste under vacuum pressure or centrifugation, while your purified PCR product is retained on the ultrafiltration membrane. Purified PCR products are recovered after a quick elution and ready for use. ExcelaPure 96-Well UF Plates can be processed manually with most vacuum manifolds and centrifuges on the market or automated for high throughput PCR purification on standard liquid handling instruments.

Kit Components	36181	95674
ExcelaPure 96-Well UF Plate	10 plates (10 x PN 4050208)	50 plates (50 x PN 4050208)

Equipment and Materials Required

1. Vacuum Manifold[1] or Variable speed centrifuge (benchtop or floor model)
2. Rotor and Microplate carriers (for centrifuge use only)
3. Deionized water or 1X TE
4. 96-Well Receiver Plate[2]
5. Multi-channel pipettor

Storage Condition

Store at Room Temperature.

Quality Control

Tested for primer removal and dsDNA recovery of different size fragments.

Recommended Vacuum Protocol for >300bp PCR Products

1. Prepare the vacuum manifold[1] according to manufacturer's instructions.
2. Place ExcelaPure 96-Well UF Plate on top of the vacuum manifold.
3. Transfer PCR products carefully to the membrane of the ExcelaPure 96-Well UF Plate.
   - Note: If the reaction volume is <100µl, add deionized water so that the final volume is 100µl.
   - Unused wells of the plate do not have to be sealed when vacuum is applied.
4. Apply vacuum[3] at 20 inches Hg for 5-10 minutes or until the wells are dry. Turn vacuum off.
   - The wells of the plate may appear shiny when dry.
   - Vacuum times increase when processing >100µl volumes.
5. Optional: Add 100µl deionized water and apply vacuum[3] at 20 inches Hg until the wells are dry. Turn vacuum off.
   - This optional washing step may be required for sensitive downstream applications.
6. Add 100µl deionized water or 1X TE.
7. Resuspend purified DNA by pipetting up and down 20 times.
   - Alternatively, mix for 10 minutes on a plate shaker. Avoid high speeds that cause displacement of samples from wells.
8. Transfer purified PCR products to a 96-Well Receiver Plate.

Recommended Vacuum Protocol for 100-300bp PCR Products

1. Prepare the vacuum manifold[1] according to manufacturer's instructions.
2. Place ExcelaPure 96-Well UF Plate on top of the vacuum manifold.
3. Transfer PCR products carefully to the membrane of the ExcelaPure 96-Well UF Plate.

- Note: If the reaction volume is < 100µl, add deionized water so that the final volume is 100µl.
- Unused wells of the plate do not have to be sealed when vacuum is applied.

4. Apply vacuum at 10 inches Hg for 10 minutes or until the wells are dry. Turn vacuum off.
   - The wells of the plate may appear shiny when dry.
   - Vacuum times increase when processing >100µl volumes.
5. Optional: Add 100µl deionized water and apply vacuum at 10 inches Hg until the wells are dry. Turn vacuum off.
   - This optional wash step may be required for sensitive downstream applications.
6. Add 100µl deionized water or 1X TE.
7. Resuspend purified DNA by vigorously pipetting up and down 20 times.
   - Alternatively, mix for 10 minutes on a plate shaker. Avoid high speeds that cause displacement of samples from wells.
8. Transfer purified PCR products to a 96-Well Receiver Plate.

Recommended Centrifugation Protocol for >100bp PCR Products

1. Stack the ExcelaPure 96-Well UF Plate on top of a suitable 96-Well Receiver Plate.
2. Transfer PCR products carefully to the membrane of the ExcelaPure 96-Well UF Plate.
   - Note: If the reaction volume is <100µl, add deionized water so that the final volume is 100µl.
   - Unused wells of the plate do not have to be sealed during centrifugation.
3. Place assembly in the centrifuge.
4. Centrifuge for 5 minutes at[4] 1400 *x g*. Discard eluate.
   - See "Additional Notes" for determination of RPM from RCF or visit our website at www.edgebio.com and click on Technical Support.
   - The wells of the plate may appear shiny when dry.
   - Centrifugation times may increase when processing >100µl volumes.
5. Optional: Add 100µl deionized water and centrifuge for 5 minutes[4] at 1400 *x g*.
   - This optional wash step may be required for sensitive downstream applications.
6. After centrifugation, add 100µl deionized water or 1X TE.
7. Resuspend purified DNA by pipetting up and down 20 times.
   - Alternatively, mix for 10 minutes on a plate shaker. Avoid high speeds that cause displacement of samples from wells.
8. Transfer purified PCR products to a 96-Well Receiver Plate.

Additional Notes

1. List of Validated Vacuum Manifolds
   - Millipore MultiScreen Vacuum Manifold Cat. No. MAVM 096 OR
   - QIAvac Multiwell Unit Cat. No. 9014597
   - Whatman UniVac 3 Vacuum Manifold Cat. No. 7705-0102
2. The following receiver plate is recommended: Nunc Polypropylene V-bottom plate (Part #442587).
3. Pressure Conversions

To convert to Inches of Mercury (in Hg) from:	Multiply by:
Millimeters of Mercury (mm Hg)	25.4
Atmospheres (atm)	0.033421
Torrs (Torr)	25.4
Millibars (mbars)	33.86
Pounds per square inch (psi)	0.491153
Kilopascals (kPa)	3.386380

4. 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 = 1400 *x g*:

$$\boldsymbol{RPM} = 35{,}400 \sqrt{\frac{1}{r}}$$