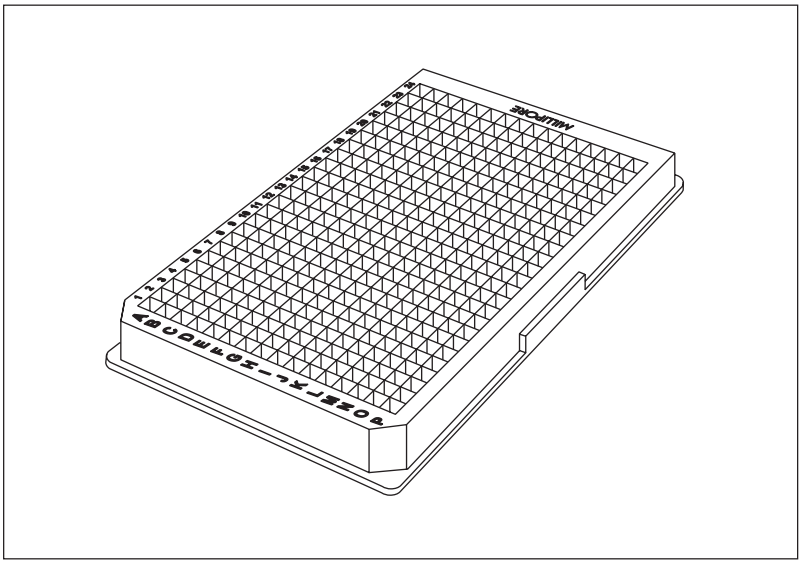
MultiScreen®$_{384}$-SEQ Plates User Guide



For research use only.
Not for use in clinical applications.
Single use only.

S384 SEQ 10 (10/pk)
S384 SEQ 50 (50/pk)

MILLIPORE

DOMINIQUE

Introduction

MultiScreen$_{384}$-SEQ plates are intended for the removal of salts and unincorporated dye terminators from sequencing reactions prior to DNA sequencing. The protocol is fast, easy to perform, and has proven effective in removing both major types of commercially available energy transfer dyes (BigDye® Terminators and DYEnamic® ET Terminators). Filtering the wells of the plate to dryness purifies the sequencing products by washing salts and unincorporated dye terminators to waste. The purified sequencing products are then resuspended from the membrane surface and are ready for injection into a capillary or slab gel type DNA Sequencer.

Protocol Guidelines

- Complete filtration is required for sample purity. The filters appear shiny even after they are dry. Continue filtration for 15–30 seconds after the last well appears empty.
- Filtration time will vary depending on the type of sequencing reactions being purified.
- Do not exceed the recommended vacuum pressure (25" Hg).

Procedure

The filtration times stated in the procedure below were developed using 1/8th BigDye reactions.

1. After thermal cycling, adjust sequencing reaction volume to 20 µL with 0.3 mM EDTA, pH 8.0 made in Milli-Q® grade water in the 96-well or 384-well thermal cycling plate. Mix gently by pipetting up and down 3–5 times.
2. Transfer all of the sequencing reaction from the thermal cycling plate to the MultiScreen$_{384}$-SEQ plate.
3. Place the MultiScreen$_{384}$-SEQ plate on the vacuum manifold. Set the vacuum pump to 23"–25" Hg and apply the vacuum for 3–4 minutes, or until the wells are completely empty. Keep applying the vacuum for an additional 15–30 seconds after the last well is empty.
4. Shut off the vacuum source and remove the MultiScreen$_{384}$-SEQ plate from the manifold.
5. Blot the excess liquid from the bottom of the MultiScreen$_{384}$-SEQ plate by briefly pressing the plate on an absorbent material such as paper towels.
6. Place the MultiScreen$_{384}$-SEQ plate back on the vacuum manifold. Add 25 µL of 0.3 mM EDTA, pH 8.0 to every well of the MultiScreen$_{384}$-SEQ plate.
7. Apply vacuum at 23"–25" Hg until the solution has been completely removed from the wells (3–4 minutes). Continue to apply vacuum for 15 to 30 seconds after the last well is empty.
8. Shut off the vacuum source and remove the MultiScreen$_{384}$-SEQ plate from the manifold.
9. Blot the excess liquid from the bottom of the plate by briefly pressing the plate on an absorbent material such as paper towels.
10 To resuspend the sequencing products, add 20 µL of 0.3 mM EDTA, pH 8.0 and pipette up and down for at least 20 cycles with a liquid handler.
11. Transfer the purified sequencing products to an appropriate plate for electrokinetic injection.

    To inject samples onto the sequencer using a high concentration formamide buffer (> 20% Formamide):

    Resuspend the samples in Milli-Q grade water and transfer them to an appropriate plate for loading onto the sequencer. Speedvac to dryness and then resuspend the samples in the formamide buffer. The samples are now ready for loading onto a gel or for electrokinetic injection.
12. Samples should be injected at 2 kV for 15–30 seconds into the ABI Prism® 3700 sequencer or at 2 kV for 30–40 seconds onto the MegaBace® 1000 sequencer. For the 3730, 3730XL, 3100 and 3100 Avant instruments, use the injection parameters supplied by ABI.

Protocol Preview

Specifications

SBS/ANSI compliant

Maximum well volume: 130 µL

Working well volume: 100 µL

Recommended vacuum pressure: 25" Hg

Distance from bottom of the plate to membrane surface: 3 mm

Materials of Construction

Plastic housing: Clear polystyrene

Membrane: Proprietary

Product Ordering Information

This section lists the catalogue numbers for MultiScreen$_{384}$ plates and equipment. See the Technical Assistance section for information about contacting Millipore. You can also buy Millipore products on-line at www.millipore.com/purecommerce.

Description	Quantity	Catalogue No.
MultiScreen$_{384}$-SEQ plate	10/pk	S384 SEQ 10
MultiScreen$_{384}$-SEQ plate	50/pk	S384 SEQ 50
MultiScreen$_{384}$-PCR plate	10/pk	S384 PCR 10
MultiScreen$_{384}$-PCR plate	50/pk	S384 PCR 50
Vacuum Manifold for 384 well plate, includes base, tubing, gauges, and flow control.	1/pk	SAVM 384 01
Chemical Duty Pump (115 V, 60 Hz)	1/pk	WP61 115 60
Chemical Duty Pump (230 V, 50 Hz)	1/pk	WP61 220 50
Chemical Duty Pump (100 V, 50/60 Hz)	1/pk	WP61 100 60
Tubing, for vacuum use; silicone, 3/16" I.D., 1.4 m	1/pk	XX71 000 04
Vacuum filtering flask, 1L	1/pk	XX10 047 05
Stopper, No. 8, perforated, silicone	1/pk	XX10 047 08
Millex®-FG$_{50}$ filter device	10/pk	SLFG 050 10

DOMINIQUE

Technical Assistance

For more information, contact the Millipore office nearest you. In the U.S., call **1-800-MILLIPORE** (1-800-645-5476). Outside the U.S., see your Millipore catalogue for the phone number of the office nearest you or go to our web site at www.millipore.com/offices for up-to-date worldwide contact information. You can also visit the tech service page on our web site at www.millipore.com/techservice.

Standard Warranty

Millipore Corporation ("Millipore") warrants its products will meet their applicable published specifications when used in accordance with their applicable instructions for a period of one year from shipment of the products. **MILLIPORE MAKES NO OTHER WARRANTY, EXPRESSED OR IMPLIED. THERE IS NO WARRANTY OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE.** The warranty provided herein and the data, specifications and descriptions of Millipore products appearing in Millipore's published catalogues and product literature may not be altered except by express written agreement signed by an officer of Millipore. Representations, oral or written, which are inconsistent with this warranty or such publications are not authorized and if given, should not be relied upon.

In the event of a breach of the foregoing warranty, Millipore's sole obligation shall be to repair or replace, at its option, the applicable product or part thereof, provided the customer notifies Millipore promptly of any such breach. If after exercising reasonable efforts, Millipore is unable to repair or replace the product or part, then Millipore shall refund to the customer all monies paid for such applicable product or part. **MILLIPORE SHALL NOT BE LIABLE FOR CONSEQUENTIAL, INCIDENTAL, SPECIAL OR ANY OTHER INDIRECT DAMAGES RESULTING FROM ECONOMIC LOSS OR PROPERTY DAMAGE SUSTAINED BY ANY CUSTOMER FROM THE USE OF ITS PRODUCTS.**

UE DUTSCHER SAS

MILLIPORE

©2004 Millipore Corporation. All rights reserved. Printed in Ireland.

Millipore, Millex, Milli-Q, and MultiScreen are registered trademarks of Millipore Corporation.
ABI Prism and BigDye are trademarks of Applera Corporation.
MegaBace is a trademark of Amersham Biosciences AB.
DYEnamic is a trademark of Amersham Biosciences UK Limited.

PR01811, Rev. –, 11/04