INSTRUCTIONS

PVDF Transfer Membrane



Pub. No. MAN0011523

88518 88585 88520

Rev. B.0 Pub Part No. 2161520.4

Number Description

88520	PVDF Transfer Membrane, 0.2µm, 26.5cm x 3.75m, 1 roll
88518	PVDF Transfer Membrane, 0.45µm, 26.5cm x 3.75m, 1 roll
88585	PVDF Transfer Membrane, 0.45µm, 10 x 10cm, 10 sheets

Storage: Store membranes flat at ambient temperature, away from chemical vapors. Some solvent vapors may partially dissolve the membranes, which will disrupt the pore structure.

Introduction

Polyvinylidene difluoride (PVDF) membranes are hydrophobic and have high binding affinity for proteins and nucleic acids. These membranes are typically used for applications such as Western, Southern, Northern and dot blots. PVDF membranes offer a better retention of adsorbed proteins than other supports including nitrocellulose.

Example Procedure for Transferring Proteins to a PVDF Membrane

Note: Use a sharp instrument to cut the membrane to the size of the gel. Any small tear may result in a larger tear. Always wear gloves when handling PVDF membranes because oils from fingers may prevent proper wetting. Proteins from hands may also bind to the membranes causing background.

A. Materials required

- Tank transfer system with power supply .
- Blotting paper (Part No. 88600)
- Methanol
- NovexTM Tris-Glycine Transfer Buffer (25x) (Part No. LC3675) or other suitable transfer buffer (e.g., 5.8g Tris base, 14.4g glycine, 0.4g SDS (optional), 200mL methanol (100%), pH 8.5. Adjust volume to 1L with ultrapure water).

B. Method

- Remove gel from the electrophoresis apparatus and equilibrate in Transfer Buffer for 30 minutes with gentle shaking. 1. Note: Incubation time is based on a 1.5mm thick gel. Reduce incubation time for thinner gels.
- 2. Cut membrane to the same dimensions of the gel. Cut a notch in the membrane corner to correspond to a corner of the gel.
- Wet membrane in 100% methanol for 15 seconds. Ensure that there are no dry areas on the membrane that could inhibit 3. protein transfer.
- 4. Place membrane in a new container with Transfer Buffer and equilibrate for 15 to 20 minutes.
- Wet the absorbent filter paper in Transfer Buffer. 5.
- Use the following component order to form the transfer stack: 6.





7. Connect the leads and perform transfer for 45-90 minutes at 0.8mA/cm² of gel.

Note: Transfer time and efficiency will vary depending upon polyacrylamide concentration, gel thickness, the presence of SDS or methanol, pH and ionic strength of the transfer buffer and the molecular weight of the protein. Determine optimal transfer conditions empirically.

- 8. When the transfer is complete, disconnect leads and disassemble the transfer stack to remove the membrane.
- 9. Keep membrane moist until ready to use.

Related Products

Novex TM Tris-Glycine protein gels (see <u>thermofisher.com/proteingels</u> for a complete listing)
Bolt TM Bis-Tris Plus protein gels (see <u>thermofisher.com/proteingels</u> for a complete listing)
iBright™ Prestained Protein Ladder
Pierce TM Reversible Protein Stain Kit for PVDF Membranes, sufficient material for 10 mini blots
Western Blotting Filter Paper, 8cm × 10.5cm, 100 sheets
SuperSignal [™] West Pico Plus Chemiluminescent Substrate, 500mL
SuperSignal West Dura Extended Duration Substrate, 100mL
SuperSignal West Femto Maximum Sensitivity Substrate, 100mL
Restore TM Western Blot Stripping Buffer, 500mL
CL-XPosureTM Film (5'' \times 7''), 100 sheets
CL-XPosure Film (8''×10''), 100 sheets
Pierce Background Eliminator Kit, for eliminating background from X-ray film

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