

ELECTROPHORESIS AND BLOTTING

Transferring High Molecular Weight Proteins on the Trans-Blot® Turbo™ Transfer System

Quick Start Guide

Many factors affect transfer of proteins from a gel to a membrane. When optimizing Trans-Blot Turbo transfers for proteins larger than 150 kD, try the following:

Prepacked Consumables

- Ready-to-use transfer packs eliminate extra membrane, filter paper, and buffer preparation. Setup time is reduced to 1 minute from the opening of the gel cassette to the start of the transfer
- Ready-to-assemble transfer kits provide all consumables to transfer 40 blots, including transfer buffer, transfer stacks, and the option to select from nitrocellulose, PVDF, and LF PVDF membranes



1 Gel Selection

- Ensure the gel is not being equilibrated in water or buffer prior to transfer. This may inhibit transfer efficiency
- Use a 4–15% or 4–20% gradient gel. For best results use Bio-Rad's TGX™ and TGX Stain-Free™ precast gels. When running the sample use the recommended conditions for each gel. These gels leave large proteins in more porous parts of the matrix, allowing for better transfer out of the gel

See reverse side for Bio-Rad's stain-free precast gel options.

2 Membrane Selection

- Nitrocellulose and PVDF have varying binding efficiencies as a result of the physiochemical properties of the membrane and the biochemical properties of the protein sample
- Try switching membrane type to improve transfer efficiency. PVDF usually performs best due to its higher binding capacity

See reverse side for membrane options for the Trans-Blot Turbo system.

3 Protocol Selection

- Select the High Molecular Weight protocol on the Trans-Blot Turbo system, not the standard Turbo protocol. The extra 3 minutes in this protocol help move large proteins through the gel matrix and onto the membrane
- Program a custom protocol if the High Molecular Weight protocol does not yield acceptable results

Note: Suggested customer protocols include



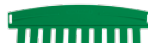

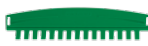

- i. For midi or 2 minis, program 2.5 A constant for 15 min
- ii. For 1 mini, program 1.3 A constant for 15 min

When checking transfer efficiency, it is common to use a colored molecular weight standard. This crude check is not always representative of the proteins in a sample. For an accurate measure of transfer efficiency, use a total protein stain or Bio-Rad's stain-free technology to obtain a true representation of how well the proteins of interest have transferred to the membrane.








Ordering Information

TGX Stain-Free Precast Gels

Description	 8+1-Well 30 µl	 10-Well 30 µl	 10-Well 50 µl	 12-Well 20 µl	 15-Well 15 µl	 IPG Well 7 cm IPG Strip
Mini-PROTEAN® TGX Stain-Free™ Precast Gels						
7.5% Resolving Gel	456-8029	456-8023	456-8024	456-8025	456-8026	456-8021
10% Resolving Gel	456-8039	456-8033	456-8034	456-8035	456-8036	456-8031
12% Resolving Gel	456-8049	456-8043	456-8044	456-8045	456-8046	456-8041
4–15% Resolving Gel	456-8089	456-8083	456-8084	456-8085	456-8086	456-8081
4–20% Resolving Gel	456-8099	456-8093	456-8094	456-8095	456-8096	456-8091
8–16% Resolving Gel	456-8109	456-8103	456-8104	456-8105	456-8106	456-8101
Any kD Resolving Gel	456-8129	456-8123	456-8124	456-8125	456-8126	456-8121

All formats are available in 10-packs (catalog numbers listed) or 2-packs (add an "S" to the end of the catalog number listed).

Description	 12+2-Well* 45 µl	 18-Well 30 µl	 26-Well 15 µl	 Prep+2-Well* 800 µl	 IPG+1-Well* 11 cm IPG Strip
Criterion™ TGX Stain-Free™ Precast Gels**					
7.5% Gel	567-8023	567-8024	567-8025	—	—
10% Gel	567-8033	567-8034	567-8035	—	—
12% Gel	567-8043	567-8044	567-8045	—	—
18% Gel	567-8073	567-8074	567-8075	567-8072	567-8071
4–15% Gel	567-8083	567-8084	567-8085	567-8082	567-8081
4–20% Gel	567-8093	567-8094	567-8095	567-8092	567-8091
8–16% Linear Gradient	567-8103	567-8104	567-8105	567-8102	567-8101
10–20% Linear Gradient	567-8113	567-8114	567-8115	567-8112	567-8111
Any kD Gel	567-8123	567-8124	567-8125	567-8122	567-8121

* Reference wells accommodate 15 µl of markers/standards. ** Criterion TGX Stain-Free gels are sold as a single gel.

Catalog #	Description
Protein Standards	
161-0373	Precision Plus Protein™ All Blue Standards
161-0363	Precision Plus Protein Unstained Standards
161-0385	Precision Plus Protein™ WesternC™ Pack
Buffers	
161-0732	10x Tris/Glycine/SDS, 1 L
161-0747	4x Laemmli Sample Buffer, 10 ml
Electrophoresis Cell	
165-6001	Criterion™ Cell, includes electrophoresis buffer tank, lid with power cables, 3 sample loading guides
165-8004	Mini-PROTEAN® Tetra Cell for Mini Precast Gels, 4-gel vertical electrophoresis system, includes electrode assembly, companion running module, tank, lid with power cables, mini cell buffer dam
Blotting System	
170-4155	Trans-Blot Turbo Starter System, blotting instrument, includes base, 2 cassettes to hold 1–2 midi or up to 4 mini blotting sandwiches, blot roller, and starter consumable kit
170-4156	Trans-Blot Turbo Transfer Pack, mini, PVDF, pkg of 10
170-4157	Trans-Blot Turbo Transfer Pack, midi, PVDF, pkg of 10
170-4158	Trans-Blot Turbo Transfer Pack, mini, nitrocellulose, pkg of 10
170-4159	Trans-Blot Turbo Transfer Pack, midi, nitrocellulose, pkg of 10
170-4270	Trans-Blot Turbo RTA Transfer Kit, mini, nitrocellulose
170-4271	Trans-Blot Turbo RTA Transfer Kit, midi, nitrocellulose
170-4272	Trans-Blot Turbo RTA Transfer Kit, mini, PVDF
170-4273	Trans-Blot Turbo RTA Transfer Kit, midi, PVDF
170-4274	Trans-Blot Turbo RTA Transfer Kit, mini, LF PVDF
170-4275	Trans-Blot Turbo RTA Transfer Kit, midi, LF PVDF

Tips for good quantitative western blotting are available at www.bio-rad.com, www.bio-rad.com/lit/2895, and www.bio-rad.com/tech/westernblotdoctor.

Catalog #	Description
Imaging Systems	
170-8195	Gel Doc™ XR+ System with Image Lab™ Software, PC or Mac, includes darkroom, UV transilluminator, epi-white illumination standard filter, camera, cables, Image Lab software
170-8265	ChemIDoc™ XRS+ System with Image Lab Software, PC or Mac, includes darkroom, UV transilluminator, epi-white illumination standard filter, camera, power supply, cables, Image Lab software
Detection Reagents	
170-5060	Clarity™ Western ECL Substrate, 200 ml
170-5061	Clarity Western ECL Substrate, 500 ml
ChemIDoc MP Imaging System	
170-8280	ChemIDoc MP Imaging System with Image Lab Software, PC or Mac, includes darkroom, UV transilluminator, epi-white illumination, camera, power supply, cables, Image Lab software
Accessories	
170-8283	Red LED Molecular Kit, pkg of 2 epi-red LED modules and 1 red emission filter, for use with applications requiring red fluorophore detection
170-8284	Green LED Molecular Kit, pkg of 2 epi-green LED modules and 1 green emission filter, for use with applications requiring green fluorophore detection
170-8285	Blue LED Molecular Kit, pkg of 2 epi-blue LED modules and 1 blue emission filter, for use with applications requiring blue fluorophore detection

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