Bio-Rad® **V3** Western Workflow™

Save time, identify problems earlier, and have more confidence in your western blot results.

Compared to conventional methods, this improved workflow, incorporating stain-free gel technology, saves time and increases the accuracy and reliability of your western blot results.

Separate Proteins Run gels in as little as 15 min Speed with flexibility: TGX Stain-Free™ Gel chemistry available in precast and handcast Visualize Protein Separation Visualize separation for all lanes in 1 min Coomassie-like performance with no background variability and no staining/destaining Stain-free image of pretransferred gel **Transfer** Efficient and uniform protein transfer in 3 min • Throughput: transfer 4 mini-gels at once **Verify Transfer Efficiency Quickly assess transfer efficiency** Verify quality of transfer for all lanes in 2 min Stain-free image of blot Validate Western Blot Data by Use stain-free blot image as total protein

Normalization and Analysis







Detect protein of interest



Normalize protein of interest with stain-free image of blot from step 4

loading control

- No need to strip and reprobe or cut the blot
- Use the entire protein sample in one lane; not dependent on a single housekeeping protein
- Reliable and accurate quantitation



FAQs

How does stain-free technology work?

The embedded compound in stain-free gels is activated and imaged, post-electrophoresis, by a Bio-Rad imager to provide a fluorescent signal with sensitivity equivalent to Coomassie staining. The stain-free signal transfers with the proteins during blotting, which enables the assessment of transfer efficiency and provides total protein signal for use in normalization.

How do total protein stains compare to normalizing with a housekeeping protein?

Advantages of total protein stains vs. housekeeping normalization include:

- Housekeeping protein (HKP) expression may change depending on experimental conditions and sample type.
 Total protein is more stable and less prone to change with experimental conditions.
- 2. Total protein normalization using stain-free technology eliminates the need to strip and reprobe for housekeeping proteins. Stripping and reprobing for HKP is a very time consuming process, adding hours to detection, whereas stain-free total protein detection can be done in minutes.
- 3. Compared to housekeeping proteins, total protein stains exhibit superior linearity and reproducibility.

How does stain-free fluorescence compare to other total protein stains for blots?

Stain-free technology provides higher detection sensitivity and a wider linear dynamic range than available total protein stains. Stain-free technology eliminates staining and destaining manipulation, ensuring proteins remain intact from gel to membrane. Other total protein stains, including Ponceau S and SYPRO Ruby, can introduce errors in the staining/destaining procedure.

What resources are available?

Tips on normalization with stain-free technology are available at Bio-Rad.com.

Bulletin 6434	Western Blot Normalization Using Image Lab™ Software
Bulletin 6390	General V3 Western Workflow Blotting Protocol
Bulletin 6360	A Method for Greater Reliability in Western Blot Loading Controls: Stain-Free Total Protein Quantitation
Bulletin 6351	V3 Stain-Free Technology Publications

Tips for good quantitative western blotting are available at Bio-Rad.com.

Bulletin 2895 Protein Blotting Guide bio-rad.com/tech/westernblotdoctor

Ordering Information

Catalog # Description

Protoin	Standarde	

1610373	Precision Plus Protein [™] All Blue Standards
1610363	Precision Plus Protein Unstained Standards

Buffers

1610732 **10x Tris/Glycine/SDS** 1610747 **4x Laemmli Sample Buffer**

Electrophoresis Cell

1656001	Criterion [™] Cell, includes electrophoresis buffer tank, lid with
	power cables, 3 sample loading guides
1658004	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 huffer dam

Blotting System

17001917	Trans-Blot® Turbo™ Transfer Starter System, mini, PVDF
17001919	Trans-Blot Turbo Transfer Starter System, midi, PVDF
17001918	Trans-Blot Turbo Transfer Starter System, mini, nitrocellulose
17001915	Trans-Blot Turbo Transfer Starter System, midi, nitrocellulose
1704156	Trans-Blot Turbo Transfer Pack, mini, PVDF, pkg of 10
1704157	Trans-Blot Turbo Transfer Pack, midi, PVDF, pkg of 10
1704158	Trans-Blot Turbo Transfer Pack, mini, nitrocellulose, pkg of 10
1704159	Trans-Blot Turbo Transfer Pack, midi, nitrocellulose, pkg of 10
1704270	Trans-Blot Turbo RTA Transfer Kit, mini, nitrocellulose
1704271	Trans-Blot Turbo RTA Transfer Kit, midi, nitrocellulose
1704272	Trans-Blot Turbo RTA Transfer Kit, mini, PVDF
1704273	Trans-Blot Turbo RTA Transfer Kit, midi, PVDF
1704274	Trans-Blot Turbo RTA Transfer Kit, mini, LF PVDF
1704275	Trans-Blot Turbo RTA Transfer Kit, midi, LF PVDF

Imaging Systems

17001401 ChemiDoc™ Imaging System 17001402 ChemiDoc MP Imaging System 1708265 ChemiDoc XRS+ System

Detection Reagents

 1705060
 Clarity™ Western ECL Substrate, 200 ml

 1705061
 Clarity Western ECL Substrate, 500 ml

 1705062
 Clarity Max™ Western ECL Substrate, 100 ml

TGX Stain-Free Precast Gels

	·		111111111		£	
	8+1-Well	10-Well	10-Well	12-Well	15-Well	IPG Well
Description	30 µl	30 µl	50 µl	20 µl	15 µl	7 cm IPG Strip
Mini-PROTEAN TGX	Stain-Free Pr	ecast Gels				
7.5% Resolving Gel	4568029	4568023	4568024	4568025	4568026	4568021
10% Resolving Gel	4568039	4568033	4568034	4568035	4568036	4568031
12% Resolving Gel	4568049	4568043	4568044	4568045	4568046	4568041
4-15% Resolving Gel	4568089	4568083	4568084	4568085	4568086	4568081
4-20% Resolving Gel	4568099	4568093	4568094	4568095	4568096	4568091
8-16% Resolving Gel	4568109	4568103	4568104	4568105	4568106	4568101
Any kD™ Resolving Gel	4568129	4568123	4568124	4568125	4568126	4568121

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

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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	5678023	5678024	5678025	_	_			
10% Gel	5678033	5678034	5678035	_	_			
12% Gel	5678043	5678044	5678045	_	_			
18% Gel	5678073	5678074	5678075	5678072	5678071			
4-15% Gel	5678083	5678084	5678085	5678082	5678081			
4-20% Gel	5678093	5678094	5678095	5678092	5678091			
8-16% Linear Gradient	5678103	5678104	5678105	5678102	5678101			
10-20% Linear Gradient	5678113	5678114	5678115	5678112	5678111			
Any kD Gel	5678123	5678124	5678125	5678122	5678121			

^{*} Reference wells accommodate 15 µl of markers/standards.

SYPRO is a trademark of Life Technologies Corporation.

Precision Plus Protein Standards are sold under license from Life Technologies Corporation, Carlsbad, CA for use only by the buyer of the product. The buyer is not authorized to sell or resell this product or its components.

Clarity Max Western ECL Substrate is manufactured by Cyanagen Srl and is the subject of patent application numbers US7855287, EP1950207, US9040252, AU2011202658, CA2742025, US8129136, and EP1962095, together with other equivalent granted patents and patent applications in other countries like CN102313732.

Bulletin 6427 Ver C US/EG 16-1055 0117

 $^{^{\}star\star}$ Criterion TGX Stain-Free Gels are sold as a single gel.