

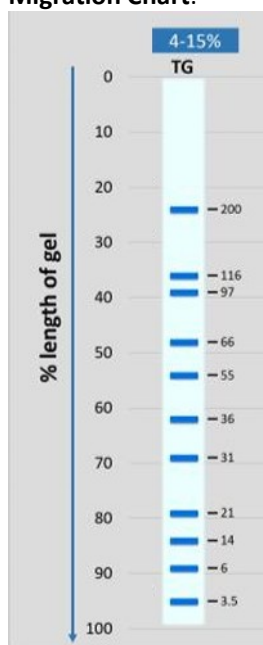
ProteoPage TGN Precast Gel (Midi, 15 wells, 4-15%)

Cat. No.: BIO-5321

Description: ProteoPage TGN Precast Gel is a convenient precast polyacrylamide gel for Tris-Glycine buffer electrophoresis systems. ProteoPage Midi gels are suitable for Invitrogen® XCell SureLock® Mini-Cell, Invitrogen® Mini Gel Tank, Hoefer SE260, and other systems.

Storage Condition: Store between 2°C - 8°C. Protect from light. Do not freeze. Keep gels flat during storage.

Migration Chart:



1.0 PROTOCOL

1.1 Recommendations prior to sample loading

- 1.1.1 Remove comb and tape before usage.
- 1.1.2 Use fresh 1X Tris-Glycine running buffer for the inner cathode chamber.
- 1.1.3 Rinse wells before loading sample.

1.2 Sample Preparation

- 1.2.1 For non-reducing samples, mix 1 part of 2X Laemmli Sample Buffer (Cat. No.: BUF-5240) to 1 part of protein sample.
- 1.2.2 For reducing samples (with 2-Mercaptoethanol), mix 1 part of 2-Mercaptoethanol to 10 parts of 2X Laemmli Sample Buffer (Cat. No.: BUF-5240). Vortex and spin down. Mix 9 parts of protein sample to above mix. Vortex and spin down. Incubate tube at 90°C for 3 minutes. Allow tube to equilibrate to room temperature.
- 1.2.3 Vortex and spin down.

1.3 Preparation of ProteoPage

- 1.3.1 Remove gel from blister packaging carefully.
- 1.3.2 Rinse gel cassette with ddH₂O.
- 1.3.3 Remove tape and comb; take extra caution not to squeeze the gel.
- 1.3.4 Insert the gel to the electrophoresis system.
- 1.3.5 Pipette-wash the wells with running buffer to remove residual storage buffer.
- 1.3.6 Load pre-stained protein markers and samples into wells.
- 1.3.7 Top-up inner and outer chamber to the highest level, ensuring that the gel wells are completely immersed in running buffer.

1.4 Recommended power settings

Voltage	150 V	200 V	250 V	300 V
Running Time* ¹	40 – 60 mins	30 – 40 mins	25 – 35 mins	15 – 25 mins
Expected current				
Initial (per gel)	40 – 50 mA	50 – 60 mA	80 – 90 mA	90 – 100 mA
Final (per gel)	10 – 20 mA	25 – 30 mA	35 – 40 mA	40 – 50 mA
Expected temperature	25 - 30°C	25 - 40°C	25 - 40°C	25- 40°C

*¹ Running time is dependent on gel percentage, running buffer freshness, temperature and power supply.

1.5 Removal of gel from cassette

Avoid gel drying. Perform gel removal immediately after electrophoresis.

- 1.5.1 Insert a cassette opener into the corners of the cassette, and gently pry the corners to separate the 2 plates.
- 1.5.2 Gently pull the plates apart from the **top** of the cassette.
- 1.5.3 Detach the gel from either the bottom or top of the cassette. Avoid peeling the gel diagonally. Use water to aid in detaching the gel if necessary.
- 1.5.4 Gently remove the gel for downstream applications.

1.6 Gel staining

- 1.6.1 ProteoPage gels can be further stained with Coomassie dyes (R-250 or G-250) and Silver-stain solution.

1.7 Protein Transfer to PVDF membrane

- 1.7.1 After detaching from cassette, equilibrate the gel in transfer buffer.
- 1.7.2 Pre-soak PVDF membrane and filter papers in transfer buffer.
 - Activate PVDF membrane in methanol before soaking in transfer buffer.
 - Prepare 6 filter papers for 1 gel sandwich.
- 1.7.3 Assemble transfer cassette in the following orientation: cathode, sponge, filter papers, gel, membrane, filter papers, sponge and anode. Protein travels in the direction of cathode to anode.
- 1.7.4 Glide roller over the transfer cassette to remove air bubbles and excess buffer until complete contact is achieved.
- 1.7.5 Insert transfer cassette into transfer module. Take note that the black side cassette should be next to the black side of the module.
- 1.7.6 Fill transfer tank with pre-cooled transfer buffer to the highest water level.
- 1.7.7 Set constant voltage at 100 V and run for 90 minutes at low temperature condition. Pre-stained protein marker should be visible on the membrane after completion of transfer.
- 1.7.8 To verify transfer of proteins, Ponceau S staining can be used before the blocking step.

2.0 Related Products

Catalog Number	Product Description
BUF-2020-10X	10X Tris Glycine (TG) Buffer
BUF-2030-10X	10X Tris Glycine-Sodium Dodecyl Sulfate (TG-SDS) Buffer
BUF-2052	20% (w/v) Sodium Dodecyl Sulfate (SDS) Solution
BUF-5210-1X	1X RIPA Buffer
BUF-5230	Acrylamide/Bis-acrylamide Solution, 30%, (37.5:1)
BUF-5231	Acrylamide/Bis-acrylamide Solution, 30%, (29:1)
BUF-5235	Acrylamide/Bis-acrylamide Solution, 40%, (37.5:1)
BUF-5236	Acrylamide/Bis-acrylamide Solution, 40%, (29:1)
BUF-5240	2X Laemmli Sample Buffer, pH 6.8
BIO-5300	ProteoBind On-column Proteolytic Digestion Kit
BIO-5305	ProteoBind Mini Column
BIO-5310	ProteoPage Bis-Tris Precast Gel (Mini, 15 wells, 4-12%)
BIO-5311	ProteoPage Bis-Tris Precast Gel (Midi, 15 wells, 4-12%)
BIO-5320	ProteoPage TGN Precast Gel (Mini, 15 wells, 4-15%)