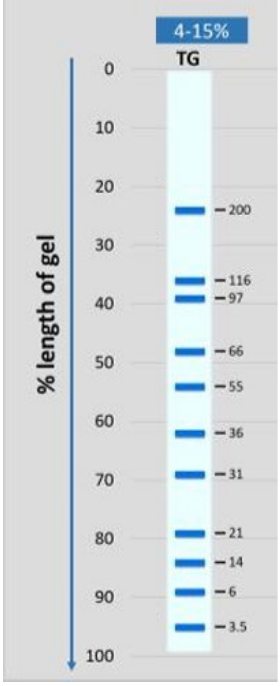
ProteoPage TGN Precast Gel (Mini, 15 wells, 4-12%)

Cat. No.: BIO-5320

Description: ProteoPage TGN Precast Gel is a convenient precast polyacrylamide gel for Tris-Glycine buffer electrophoresis systems. ProteoPage Mini gels are suitable for Bio-Rad® and common protein electrophoresis 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 ddH_2O.
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 casette 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-5320	ProteoPage TGN Precast Gel (Mini, 15 wells, 4-15%)