YamayBio

All-in-One Low Range Protein Gel Kit

QUICK START GUIDE

Research use only.
Not for diagnostic procedures.

Contents and storage

Product	Cat. No.	Quantity	Component	Cat. No.	Volume
All-in-One Low Range Protein Gel Kit (Tricine-PAGE, 10%)	RF1531	10 Assays	AIO-Stacking Buffer A	RF1551	15 mL
			AIO-Stacking Buffer B	RF1552	15 mL
			AIO-Resolving Buffer A (10%)	RF1553	30 mL
			AIO-Resolving Buffer B (10%)	RF1554	30 mL
			Enhanced Catalyst	RF1500	1 mL
			Tricine Protein Sample Loading Buffer (Denaturing, Reducing, 2×)	RF1557	3 mL
			Tricine SDS Running Buffer Instant Granules	MS8136	10 sticks (500mL×10)
			Rapid Coomassie Blue Staining (Destaining-free)	BM3151	100 mL
All-in-One Low Range Protein Gel Kit (Tricine-PAGE, 16%)	RF1532	10 Assays	AIO-Stacking Buffer A	RF1551	15 mL
			AIO-Stacking Buffer B	RF1552	15 mL
			AIO-Resolving Buffer A (16%)	RF1555	30 mL
			AIO-Resolving Buffer B (16%)	RF1556	30 mL
			Enhanced Catalyst	RF1500	1 mL
			Tricine Protein Sample Loading Buffer (Denaturing, Reducing, 2×)	RF1557	3 mL
			Tricine SDS Running Buffer Instant Granules	MS8136	10 sticks (500mL×10)
			Rapid Coomassie Blue Staining (Destaining-free)	BM3151	100 mL

Storage:

- Enhanced catalyst: store at -20 °C (stable for 12 months).
- Tricine Protein Sample Loading Buffer (Denaturing, Reducing, 2×):store at -20 °C
- Instant Granule: store at room temperature (15–25 °C).
- Other components: store at 4 °C, stable for 12 months.
- Product is shipped at ambient temperature.

After opening:

- Enhanced catalyst may be stored at 4 °C for up to 3 months.

Introduction

This kit contains a complete set of reagents required for low molecular weight protein electrophoresis (polypeptide electrophoresis). It is intended for the denaturing electrophoresis of proteins in the 2–20 kDa range and provides high resolution for the effective separation of polypeptides in the 2–5 kDa range. Each kit allows the preparation of up to 10 PAGE gels (8 × 10 cm, thickness 0.75 mm or 1 mm). The gels are free of SDS and are therefore also suitable for native electrophoresis. The kit is supplied with Tricine SDS running buffer instant granules, which eliminate the need to differentiate between anode and cathode buffers.

Quick Cast Protocol

Preparation of resolving solutions and stacking solutions for gel casting

For 1 mm thick mini-gel						
Gel Percentage	**AIO-Resolving Buffer A**	**AIO-Resolving Buffer B**	**Catalyst**	**AIO-Stacking Buffer A**	**AIO-Stacking Buffer B**	**Catalyst**
Each	2.3 mL	2.3 mL	50µL	0.75 mL	0.75 mL	15µL

For 0.75 mm thick mini-gel						
Gel Percentage	**AIO-Resolving Buffer A**	**AIO-Resolving Buffer B**	**Catalyst**	**AIO-Stacking Buffer A**	**AIO-Stacking Buffer B**	**Catalyst**
Each	1.75 mL	1.75 mL	35µL	0.5 mL	0.5 mL	10µL

For 1.5 mm thick mini-gel						
Gel Percentage	**AIO-Resolving Buffer A**	**AIO-Resolving Buffer B**	**Catalyst**	**AIO-Stacking Buffer A**	**AIO-Stacking Buffer B**	**Catalyst**
Each	3.45 mL	3.45 mL	70µL	1.00 mL	1.00 mL	20µL

*Volumes listed are sufficient for casting one 7.4 x 8.2 cm mini-gel and can be multiplied by N (desired number of gels) to cast multiple gels at once.

Instructions:

Gel preparation

1. Prepare the resolving gel with the desired acrylamide percentage by pipetting **equal volumes** of **AIO-Resolving Buffer A** and **AIO-Resolving Buffer B** into a clean conical tube.
2. Prepare the stacking gel by pipetting **equal volumes** of **AIO-Stacking Buffer A** and **AIO-Stacking Buffer B** into a clean conical tube.
3. Add the required volume of **Enhanced Catalyst** into the **resolving tube**. Gently mix reagents, avoiding the introduction of air bubbles into the gel mixture. Using a pipette, fill each cassette to 0.5-1 cm below the comb teeth.
4. Add the required volume of **Enhanced Catalyst** into the **stacking tube**. Gently mix reagents, avoiding the introduction of air bubbles into the gel mixture.
5. Position the pipette at the middle of the cassette and gently add the stacking gel, filling to the top of the short plate. A dip may occur where pipetting takes place, but it will level out.
6. Quickly and carefully insert the comb and avoid air bubble entrapment below the teeth.
7. Allow gels to polymerize for 15 minutes.
8. Gels can be used immediately or wrapped in DI water-soaked paper towels and stored in an airtight container at 4°C for up to 5 days.

Electrophoresis

1. Dissolve one stick in distilled water and adjust the final volume to 500 mL to prepare 1× Tricine SDS electrophoresis buffer.
2. Mix the sample with an **equal volume** of **Tricine sample loading buffer** (denaturing, reducing, 2×). Heat at 95 °C for 5–10 minutes, centrifuge at high speed for 5 minutes, and use the supernatant for electrophoresis analysis.
3. Fill the inner chamber of the electrophoresis tank with Tricine SDS electrophoresis buffer, and add an appropriate volume of the same buffer to the outer chamber. Carefully remove the comb and rinse the wells with a pipette. Load the prepared protein samples or marker into the wells, and perform electrophoresis under the recommended constant voltage conditions:
   - 10% Tricine gel: 120 V, 1 h
   - 16% Tricine gel: 120 V, 2 h

Note:
After electrophoresis, proceed with the subsequent steps as soon as possible to prevent diffusion of polypeptides out of the gel.

Staining

1. Since polypeptides tend to diffuse within the gel, the Tricine gel after electrophoresis may be **fixed in isopropanol fixation solution** (50% isopropanol, 10% acetic acid, 40% distilled water) for 30 minutes before staining.
2. Discard the fixation solution and add **30 mL Coomassie Brilliant Blue** rapid stain (no destaining required), ensuring the gel is fully covered. Stain on a horizontal shaker at room temperature for 10–15 minutes. After staining, discard the staining solution and rinse the gel with distilled water to remove residual dye. The results can then be observed directly.