YamayBio

All-in-One Low Range Protein Gel Kit

QUICK START GUIDE

For research use only.
Not for use in 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 (500 mL×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 (500 mL×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 Granules: Store at room temperature (15–25 °C).
- Other components: Store at 4 °C (stable for up to 12 months).
- The product is shipped at room temperature.

After opening:

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

Introduction

This kit provides a complete set of reagents for low-molecular-weight protein (polypeptide) electrophoresis. It is designed for denaturing electrophoresis of proteins in the 2–20 kDa range and offers high resolution for the effective separation of polypeptides between 2–5 kDa.
Each kit supports preparation of up to 10 PAGE gels (8 × 10 cm, 0.75 mm or 1 mm thickness). The gels are SDS-free and are therefore also suitable for native electrophoresis. Tricine SDS running buffer instant granules are supplied, eliminating the need to prepare separate 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	50 µL	0.75 mL	0.75 mL	15 µL

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	35 µL	0.5 mL	0.5 mL	10 µL

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	70 µL	1.00 mL	1.00 mL	20 µL

*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 by mixing **equal volumes** of **AIO-Resolving Buffer A** and **AIO-Resolving Buffer B** in a clean conical tube.
2. Prepare the stacking gel by mixing **equal volumes** of **AIO-Stacking Buffer A** and **AIO-Stacking Buffer B** in a clean conical tube.
3. Add the required volume of **Enhanced Catalyst** to the **resolving gel mixture**. Gently mix without introducing air bubbles into the gel mixture and fill each cassette to 0.5-1 cm below the comb teeth.
4. Add the required volume of **Enhanced Catalyst** to the **stacking gel**. Gently mix reagents without introducing air bubbles into the gel mixture.
5. Carefully 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 to avoid entrapment of air bubbles 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 of Tricine SDS Running Buffer Instant Granules in distilled water and adjust the final volume to 500 mL to prepare 1× running buffer.
2. Mix samples 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.