QUICK REFERENCE

Pub. No. MAN0009818 Rev. C.0



Contents and storage

Gel type	Amount	Storage
NuPAGE™ Tris-Acetate Gels	Box of 2 or 10 gels	Store at 2–8°C for up to 8 months.
	Box of 2 of To gets	Do not freeze.



Product description

NuPAGE™ Tris-Acetate Gels are precast polyacrylamide gels designed for optimal separation and resolution of large-sized proteins (36–500 kDa) under denaturing or native gel electrophoresis conditions.

NuPAGE™ Tris-Acetate Mini Gels are available with the following specifications:

■ **Polyacrylamide percentage**: 7% and 3–8%

• Well format: 10, 12, 15, and 2D wells

• Thickness: 1.0 mm and 1.5 mm



Required materials

- Protein sample and protein ladder
- NuPAGE[™] Sample Reducing Agent (10X) (for reduced samples)
- NuPAGE[™] Antioxidant (for reduced samples)
- Novex[™] Power Supply Adapters (Cat. No. ZA10001) if not using a Thermo Fisher Scientific[™] power supply
- Mini Gel Tank (Cat. No. A25977) or XCell SureLock™ Mini-Cell (Cat. No. EI0001)

For denaturing applications	For native applications
■ NuPAGE™ LDS Sample Buffer (4X)	■ Tris-Glycine Native Sample Buffer (2X)
 NuPAGE[™] Tris-Acetate SDS Running Buffer (20X) 	 Novex[™] Tris-Glycine Native Running Buffer (10X)



- Visit thermofisher.com/proteingels for additional information and protocols.
- For support, visit thermofisher.com/support.

Choosing a well format

Thicker 1.5 mm gels with fewer wells are recommended for large samples (>30 μ L). Thinner 1 mm gels are recommended for blotting because of better protein transfer.

Well type	Maximum loading volume [1]		Maximum	
	1 mm thickness	1.5 mm thickness	protein load	
10-well	25 μL	37 μL	0.5 μg/band	
12-well	20 μL	_	0.5 μg/band	
15-well	15 µL	25 μL	0.5 μg/band	
2D-well	400 μL	_	12 μg/band	

^[1] Not every format is available for every gel type.

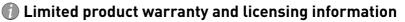
Choosing a protein ladder for your application

Туре	Marker	Cat. No.
Pre-Stained	Spectra™ Multicolor High Range Protein Ladder	26625
Pre-Stained	HiMark [™] Pre-stained Protein Standard	LC5699
Unstained	NativeMark [™] Unstained Protein Standard	LC0725
Western blot	iBright™ Prestained Protein Ladder	LC5615
Western blot	MagicMark™ XP Western Protein Standard	LC5602

Go to thermofisher.com/proteinladders for more information on protein ladders.

Choosing buffers for your application

Buffer	Application	Cat. No.
NuPAGE™ Tris-Acetate SDS Running Buffer	Resolve large-size proteins under denaturing conditions	LA0041
Tris-Glycine Native Running Buffer	Resolve large-size proteins under native (non-denaturing) conditions	LC2673
NuPAGE™ Transfer Buffer	Wet transfer	NP0006





Perform denaturing protein gel electrophoresis using NuPAGE™ Tris-Acetate Mini Gels

Step			Action				
			Prepare 1X Sample Buffer for dilutions of samples if needed. Volumes are provided for a 10-µL sample size. Scale volumes proportionally for larger sample sizes.				
			Components	Denaturing sample [1]	Native Sample		
			Sample	xμL	xμL		
			NuPAGE™ LDS Sample Buffer (4X)	2.5 µL	_		
1		Prepare samples	Tris-Glycine Native Sample Buffer (2X)	_	5 μL		
			Deionized Water	to 7.5 μL	to 5 μL		
			Total Volume	10 μL ^[2]	10 μL ^[2]		
			[1] For reduced samples, add NuPAGE™ Reducing Agent (10X) to 1X. [2] See "Choosing a well form	mat" for recommended loading volu	imes.	
			Heat denaturing samples at 70°C for 10 mir	nutes. Do not heat native sam	nples.		
2		Prepare buffers	Denaturing Buffer : Add 50 mL of 20X NuPAGE™ Tris-Acetate SDS Running Buffer to 950 mL of deionized water to prepare 1X SDS Running Buffer.				
		Trepare barrers	Native Buffer: Add 100 mL of 10X Tris-Glycine Native Running Buffer to 900 mL of deionized water to prepare 1X Native Running Buffer.				
	(🔷)		a. Remove the comb, and rinse the gel wells three times using 1X Running Buffer.				
3	Charles of the Charle	Prepare gel	b. Remove the white tape near the bottom of the gel cassettes.				
		.,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	c. Place the gels in the mini gel tank.				
			Fill the chambers with the appropriate 1X ru	inning buffer.			
		Load buffers	Mini Tank: Add 400 mL of buffer to each chamber.				
4			XCell SureLock™ Mini-Cell: Add 600 mL of buffer to the upper chamber, and 200 mL to the lower chamber (for				
			reduced samples, use running buffer with antioxidant in the lower chamber).				
	(1)	Load samples and	a. Load the appropriate volume of your samples in the appropriate wells.				
5		ladders	b. Load your protein ladder in the appropriate well.				
			· · · · · · · · · · · · · · · · · · ·				
	_		Optimal run times vary depending on gel percentage and power supply used for electrophoresis.				
		Nun the gel	For denaturing electrophoresis run for 1 hour at 150 V constant.				
6	7		For native electrophoresis run for 1.5–3 hours at 150 V constant.				
	J		Note: If you are not using a Thermo Fisher S	scientific™ power supply, insta	all Novex™ Power Supply A	dapters	
		l .					

Buffer formulation

The following recipes are provided to allow preparation of buffers from scratch. The pH listed for each buffer is for the 1X solution. **Do not use acid or base to**

The pH listed for each buffer is for the 1X solution. **Do not use acid or base to adjust the pH**. Buffers are stable for 6 months when stored at 4°C.

	Prepare 500 mL of 20X Tris-Acetate SDS Running Buffer			Pro	epare 500 mL o Run	f 10X Tris-Glyc ning Buffer	ine Native	
	50 mM Tricine, 50 mM Tris Base, 0.1% SDS, pH 8.24			25	25 mM Tris Base, 192 mM Glycine, pH 8.3			
Dissolve the following reagents in 400 mL ultrapure water.			Dissolve the following reagents in 900 mL ultrapure water.					
		Reagent	Amount					
		Tricine	89.5 g			Reagent	Amount	
		Tris Base	60.6 g			Tris Base	29 g	
						Glycine	144 g	
		SDS	10.0 g					
2. Mix well and adjust the volume to 500 mL with ultrapure water.3. Before electrophoresis, dilute buffer to 1X with water.			3.	Mix well and ad 1,000 mL with Before electrop to 1X with wate	ultrapure wate phoresis, dilute	r.		

Prepare 125 mL of 20X Transfer Buffer

25 mM Bicine, 25 mM Bis-Tris (free base), 1 mM EDTA, pH 7.2

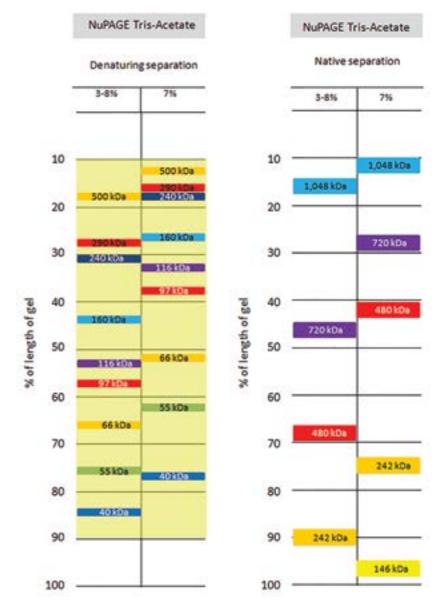
1. Dissolve the following reagents in 100 mL ultrapure water.

Reagent	Amount
Bicine	10.2 g
Bis-Tris (free base)	13.1 g
EDTA	0.75 g

- 2. Mix well and adjust the volume to 125 mL with ultrapure water.
- 3. Before western transfer, dilute buffer to 1X with water.

Migration patterns of protein standards on NuPAGE™ Tris-Acetate gels

Refer to the migration chart to find the gel best suited for your application. Your proteins of interest should migrate through ~70% of the length of the gel for the best resolution.





Choosing the right gel type for your application

Thermo Fisher Scientific protein gels					
Gel type	Gel % available	Separation range	Shelf life	Average run time	Applications
Bolt™ Bis-Tris Plus	8%, 10%, 12%, 4–12%	6 to 400 kDa	up to 16 months	22–45 min	Best choice for separation of small- to medium-sized proteins. Neutral pH environment minimizes protein modifications. Wedge well design can accommodate large sample volumes. Ideal for Western blot transfer and analysis, and all techniques in which protein integrity is crucial.
NuPAGE™ Bis-Tris	8%, 10%, 12%, 4–12%	1.5 to 300 kDa	12 months	35–50 min	Separation of small- to medium-sized proteins. Neutral pH environment minimizes protein modifications.
NuPAGE™ Tris-Acetate	7%, 3–8%	36 to 500 kDa	6 months	60 min	Separation of larger proteins.
Novex™ Tricine	10%, 16%	2 to 200 kDa	1–2 months	90 min	Separation of small proteins and peptides.
NativePAGE™ Bis-Tris	3–12%, 4–16%	15 to 10,000 kDa	12 months	90-120 min	Separation of native proteins.
Novex™ Tris-Glycine	6%, 8%, 10%, 12%, 4–20%, 4–12%, 14%, 16%, 8–16%, 10–20%	8 to 250 kDa	up to 12 months	90 min	Separation of small- to medium-sized proteins using traditional Laemmli-style gels.
E-PAGE™	E-PAGE™ 96 6% E-PAGE™ 48 8%	10 to 220 kDa	6 months	14-25 min	High-throughput for recombinant production analysis and protein profiling.
IEF	pH 3-7, pH 3-10	pl 3.5 to pl 8.5	2 months	150 min	Separation of proteins based on isoelectric point.
Zymogram	10% (gelatin)	10 to 220 kDa	2 months	90 min	Separation of proteins based on size and activity.

