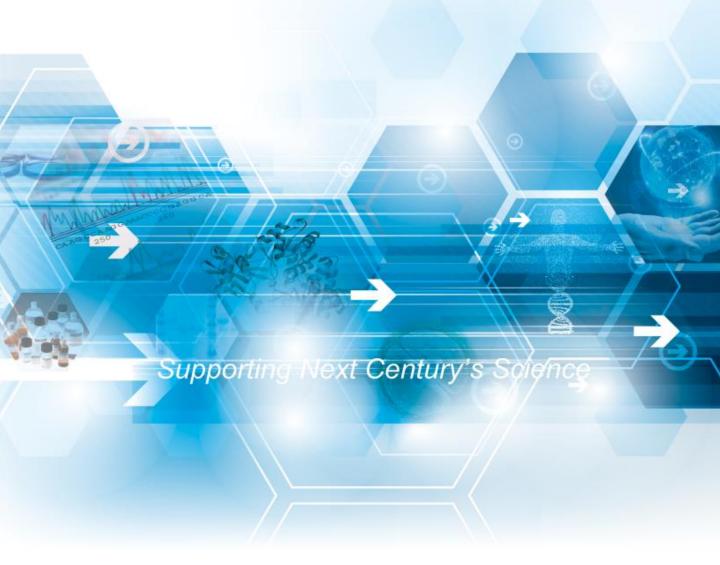
# SDS-PAGE Western Blotting





### **SDS-PAGE**

### CosmoPAGE Precast Gel

#### **Features**

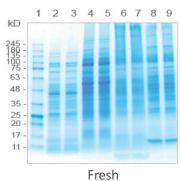
- Consistent performance
- · Compatible with Bio-Rad Mini-PROTEAN Core
- · Sharp, straight bands
- · Long shelf life; Up to 12 months

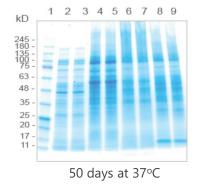




#### Consistent Performance

CosmoPAGE gels perform high protein resolution and reproducibility even stored at ambient temperature for 50 days.





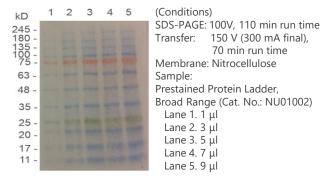
(Conditions)
Gel: CosmoPAGE TG Gel 4-15 %
Voltage: 150V for an hour
Sample:
Lane 1. Prestained Protein Ladder,
Broad Range, 3 µl
(#NU01002)
Lane 2. 3. E. coli lysate
Lane 4. 5. Hek293T lysate
Lane 6. 7. Mouse brain membrane
Lane 8. 9. Mouse brain cytosol
(10 µg each)

### Compatibility

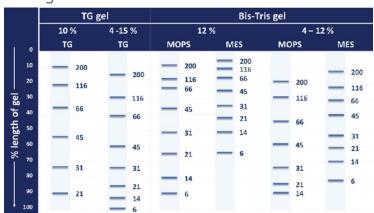
Compatible with the Bio-Rad Mini-PROTEAN Core system without use of extra adapters.



### High Transfer Efficiency



### **Protein Migration Chart**





### **Ordering Information**

Product Name	Cat. No.	PKG size	Price
CosmoPAGE TG (Tris-Glycine) Precast Gel, 10%, 15 wells	NU001015	10 gels	\$ 89.00
CosmoPAGE TG (Tris-Glycine) Precast Gel, 4-15%, 15 wells	NU041515	10 gels	\$ 89.00
CosmoPAGE Bis-Tris Precast Gel, 12%, 15 wells	NU001215	10 gels	\$ 89.00
CosmoPAGE Bis-Tris Precast Gel, 4-12%, 15 wells	NU041215	10 gels	\$ 89.00
CosmoPAGE MOPS Running Buffer, Powder	NU01004	5 x 1 L	\$ 25.00
CosmoPAGE Tris-Glycine Running Buffer, Powder	NU01011	5 x 1L	\$ 25.00
Running Buffer Solution (10x) for SDS - PAGE, Tris - Glycine	30329-61	1 L	\$ 63.00
	30329-74	5 L	\$ 231.00

# Sample Buffer Solution for SDS-PAGE (6x)

#### **Features**

- · Suitable for low concentration protein sample adjustment
- · No precipitation in the refrigerator
- · Two types; with and without reducing agent

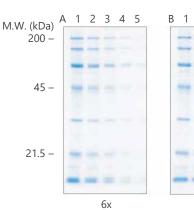
### Comparison of 6x and 2x

The data right shows that both 2x and 6x sample buffer solution have the same separation patterns. Using the Sample Beffer Solution (6x) enables reduction of valuable protein sample for SDS-PAGE.

#### (Conditions)

Prepare two types of sample buffer solutions, A: 6x and B: 2x of protein markers and separated by SDS-PAGE with 12% acrylamide gel. For staining, CBB Staining solution was used.

Amount of protein of each lane was Lane 1: 5.1  $\mu$ g, 2: 2.5 $\mu$ g, 3: 1.3  $\mu$ g, 4: 0.51  $\mu$ g and 5: 0.26  $\mu$ g.





Ordering Information

Product Name	Cat. No.	PKG size	Price
Sample Buffer Solution with Reducing Reagent (6x)	09499-14	5 ml	\$ 53.00
Sample Buffer Solution without Reducing Reagent (6x)	09500-64	5 ml	\$ 53.00
Sample Buffer Solution without 2-ME (2x)	30567-12	25 ml	\$ 59.00

#### **Related Products**

Product Name	Cat. No.	PKG size	Price
T. O. J. J. D. J.	07277-61	1 g	\$ 91.00
Tris(2-carboxyethyl)phosphine Hydrochloride [TCEP]	07277-16	10 g	\$ 405.00

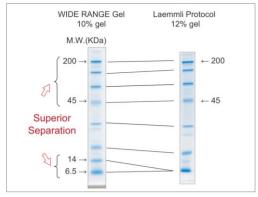


# WIDE RANGE Gel Preparation Buffer (4x) for PAGE

#### **Features**

- Simple casting procedures
   WIDE RANGE Gel Preparation buffer is a 4x concentrated neutral pH buffer. It can be used for preparation of both stacking gel and resolving gel by replacing the Tris-HCL buffer in Laemmli buffer system.
- A wide separation range
   WIDE RANGE Gel provides a much greater separation range than the gel casted with a conventional Laemmli buffer system.
- Improved stability and strength
   The increased tensile strength allows easy handling even a low percentage gel. The neutral pH buffer improves the stability of gel resulting in a longer shelf life than the gel with Laemmli buffer system.

### Comparison of Separation





### Ordering Information

Product Name	Cat. No.	PKG size	Price
WIDE RANGE Gel Preparation Buffer (4x) for PAGE	07831-94	250 ml	\$ 103.00

# Rapid Running Buffer Solution (20x)

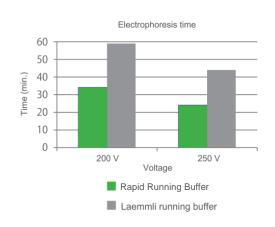
#### **Features**

- Fast Run Time: Running mini-gel in 25 minutes.
- Easy to Use: Premixed (20x) buffer just replace the Laemmli running buffer with Rapid Running buffer
- Wide-Range Separation: Gradient gel-like separation on a single percentage gel

### Comparison of electrophoresis time



YouTube: Rapid Tunning Buffer



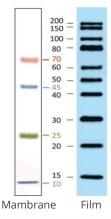
Product Name	Cat. No.	PKG size	Price
Rapid Running Buffer Solution (20x)	12981-74	250 ml	\$ 58.00

# **Protein Ladder**

### CosmoPAGE WB Ladder

#### **Features**

- 4 prestained proteins for clear visualization during electrophoresis and Western blotting transfer
- 10 IgG-binding proteins for visualization on Western blots
- · Wide range size estimation (10-200 kDa)
- · Two enhanced bands (30 and 80 kDa)

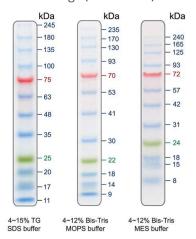


### Ordering Information

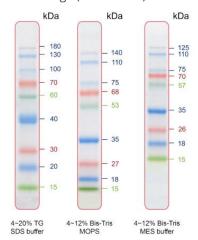
Product Name	Cat. No.	PKG size	Price
CosmoPAGE WB Ladder	NU01008	250 μΙ	\$ 160.00

# **Prestained / Unstained Protein Ladder**

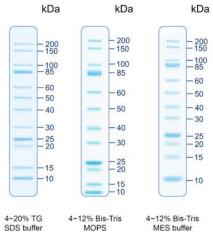
Cat. No.: NU01002 Prestained Protein Ladder, Broad Range (10-245 kDa)



Cat. No.: NU01009 Prestained Protein Ladder, Mid Range (15-180 kDa)



Cat. No.: NU01010 Unstained Protein Ladder, Broad Range (10-200 kDa)



Product Name	Cat. No.	PKG size	Price
Prestained Protein Ladder, Broad Range	NU01002	500 μΙ	\$ 100.00
Prestained Protein Ladder, Mid Range	NU01009	500 μΙ	\$ 100.00
Unstained Protein Ladder, Broad Range	NU01010	500 μΙ	\$ 100.00

CosmoPAGE WB Ladder



Prestained Protein Ladder



Unstained Protein Ladder



# **Blocking Reagents**

# **Bullet Blocking One**

#### **Features**

- Fast blocking in 5 min.
- · Ready-to-use

#### Comparison Data

Blocking Reagents	Bullet Blocking One	Blocking One	5% Skim Milk
Blocking Time	5 min.	30 min.	60 min.
Blotting Image		==	

<sup>\*</sup> Blocking One is our standard blocking reagent.

(Conditions)

Blotting: Semi-dry Blotting Buffer Solution (Cat. No.: 30650) at 10 V for 30 min

Washing: 0.1% t-TBS (Cat. No.: 12750)
Blocking: Refer to the figure above

1st antibody: Anti-Vimentin (C-20) (Rabbit) (Cat. No.: SC-7557-R) diluted 1:2,000, 1 hr. at RT

2nd antibody: Anti-Rabbit IgG-HRP (Cat. No.: SC-2004) diluted 1:100,000, 1 hr. at RT

Detection: Chemi-Lumi One (Cat. No.: 11644), 5 min. reaction time

Detector: LAS-3000 (High mode), 15 min. exposure time

Sample: 20 µg of HeLa cell extract, 5 serial two-fold dilution series

### Ordering Information

Product Name	Cat. No.	PKG size	Price
Bullet Blocking One for Western Blotting (#13779-56 x 4 bottles)	13779-14	200 ml	\$ 63.00
Bullet Blocking One for Western Blotting	13779-01	1 L	\$ 189.00

#### **Related Products**

Product Name	Cat. No.	PKG size	Price
Blocking One	03953-66	100 ml	\$ 42.00
	03953-95	500 ml	\$ 63.00
Blocking One-P for Immunoassays with Phospho-specific Antibodies	05999-84	200 ml	\$ 52.00



# **Antibody Diluent**

# Signal Enhancer HIKARI

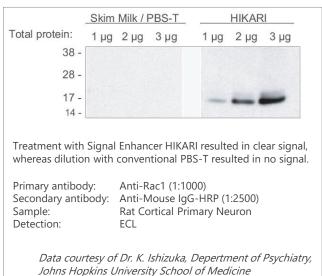
Dilute your antibodies with Signal Enhancer HIKARI instead of conventional diluents such as PBS-t or TBS-t before performing your next western blotting detection protocol and witness a remarkable increase in the ability to detect the protein of interest and to eliminate undesired background. Signal Enhancer HIKARI was developed to resolve the problems of low sensitivity and high background often encountered during procedures such as Western blotting and ELISA.

#### **Features**

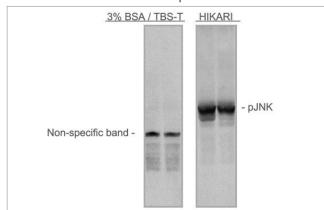
- Enhances antigen-antibody reactions -
- Removes background
- Works with any substate
- Works with any membrane
- · Ready-to-use reagent

- yield several 10-fold increase in signal intensity
- improve the specificity of your antibodies
- enhances chemiluminescence, colorimetric and fluorescence detection system
- enhances signal from nitrocellulose and PVDF membrane
- just dilute your antibodies with Signal Enhancer HIKARI

#### **Detection Enhancement of Rac1**



#### Detection Enhancement of p-JNK



Treatment with Signal Enhancer HIKARI resulted in clear signal enhancement and background suppression compared with the conventional method using TBS-T.

Primary antibody: Anti-pJNK (1:1000)

Secondary antibody: Anti-Rabbit IgG-HRP (1:5000)
Sample: Mouse Embryonic Fibroblast (MEF)

Detection: Super Signal West Pico

Data courtesy of Dr. S. Matsuzawa, Signal Transduction, NCI Cancer Center, Burnham Institute for Medical Research

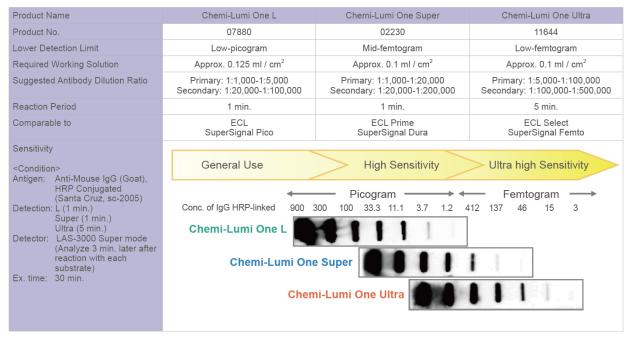
Product Name	Cat. No.	PKG size	Price
Signal Enhancer HIKARI 50 (Solution 1* 50 ml, Solution 2** 50 ml)	NU00101	1 kit	\$ 98.00
Signal Enhancer HIKARI 250 (Solution 1* 250 ml, Solution 2** 250 ml)	NU00102	1 kit	\$ 298.00
Signal Enhancer HIKARI - Solution 1	NU00102-1	250 ml	\$ 179.00
Signal Enhancer HIKARI - Solution 2	NU00102-2	250 ml	\$ 179.00



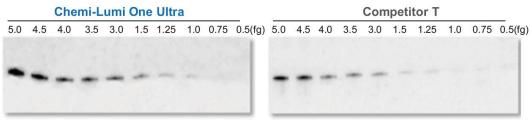
### **Chemiluminescent HRP Substrates**

### **Chemi-Lumi One Series**

Chemi-Lumi One is a series of highly sensitive luminol-based chemiluminescence assay kits for Western Blotting. Three types of chemiluminescent substrates are available for Western blot detection with horseradish peroxidase enzyme (HRP).



#### Chemi-Lumi One Ultra



Primary antibody: Rabbit anti-transferrin (1: 30,000) Secondary antibody: Goat-anti-rabbit IgG-HRP (1: 200,000)

Product Name	Cat. No.	PKG size	Price
Chemi-Lumi One L (Luminol 50 mL, Peroxide 50 mL)	07880-54	800 cm <sup>2</sup>	\$ 84.00
Chemi-Lumi One L (Luminol 250 mL, Peroxide 250 mL)	07880-70	4,000 cm <sup>2</sup>	\$ 200.00
Chemi-Lumi One Super (Luminol 10 mL, Peroxide 10 mL)	02230-14	200 cm <sup>2</sup>	\$ 84.00
Chemi-Lumi One Super (Luminol 50 mL, Peroxide 50 mL)	02230-30	1,000 cm <sup>2</sup>	\$ 210.00
Chemi-Lumi One Ultra (Luminol 10 mL, Peroxide 10 mL)	11644-24	200 cm <sup>2</sup>	\$ 116.00
Chemi-Lumi One Ultra (Luminol 50 mL, Peroxide 50 mL)	11644-40	1,000 cm <sup>2</sup>	\$ 328.00







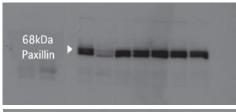
# **Stripping and Reprobing**

# **WB Stripping Solution**

#### **Features**

- No heating
- Reaction at room temperature
- No odor
- Does not contain 2-mercaptoethanol
- Fast
- Stripping time 5-15 minutes
- Ready-to-use
- One solution in one bottle

#### **Applications**





First antigen-antibody reaction

Blocking: Blocking One (Cat. No.: 03953-95), 30 min

Wash: t-Tris Buffered Saline
Primary antibody: Anti-Paxillin (mouse IgG)
Secondary antibody: Anti-mouse IgG-POD

Detection: Chemiluminescence Detection Kit (commercially available product)

Stripping

RT, 15 min. for conventional protocol

Second, different antigen-antibody reaction

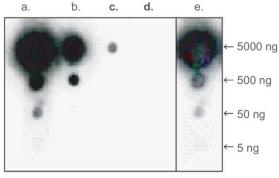
Blocking: Blocking One (Cat. No.: 03953-95), 30 min

Wash: t-Tris Buffered Saline

Primary antibody: Anti-Vinculin (mouse IgG) / Anti-Actin (mouse IgG)

Secondary antibody: Anti-mouse IgG-POD Detection: Chemi-Lumi One

# Comparison of WB Stripping Solution an WB Stripping Solution Strong



Apply HPR-labeled anti-GST antibody to 5000 ng, 500 ng, 50 ng, or 5 ng (as desired) of c-Myc-GST antigen on a PVDF membrane, then remove the antibody by agitating gently for 10 minutes using one of the following stripping solutions.

- a: 0.05%(v/v) t-TBS
- b: 2%(w/v) SDS, 100mM 2-Mercaptoethanol
- c: WB Stripping Solution
- d: WB Stripping Solution Strong

After stripping the antibodies and washing the membrane with t-PBS for 2 min, use the chemiluminescence method to detect the HPR-labeled anti-GST antibody remaining on the membrane.

\*Image "e" is a result that shows detection of the antigen with HPR-labeled anti-GST antibody on the "d" . The similar result is marked with "a" . Therefore, WB Stripping Solution Strong only stripped antibodies, not antigens.

Product Name	Cat. No.	PKG size	Price
WB Stripping Solution	05364-55	500 ml	\$ 98.00
WB Stripping Solution Strong	05677-65	500 ml	\$ 180.00
WB Stripping Solution Trial Set (WB Stripping Solution + WB Stripping Solution Strong)	05680-21	40 ml each	\$ 30.00



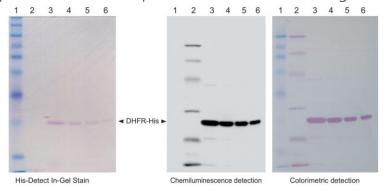
# **Staining His-tagged Fusion Protiens**

### His-Detect In-Gel Stain

#### **Features**

- · Stain His-tagged fusion proteins directly in gels without western blotting
- Detect picomolar level of His-tagged fusion proteins within an hour
- · No fluorescent image analyzer or transilluminator required

#### Application Data: Comparison with western blotting



Western blotting: Primary antibody: Anti-6xHis (Cat. No.: 04428) Secondary antibody: Anti-Mouse IgG-HRP (Goat) (SantaCruz, Cat. No.: SC-2005)

Cahemiluminescence detection: Chemi-Lumi One L (Cat. No.: 07880) Colorimetric detection: Peroxidase Stain Kit (Cat. No.: 26652)

\* For detection of Protein Markers (Biotinylated), Streptavidin Horseradish Peroxidase Conjugate (Cat. No.: 02517) is used.

#### Ordering Information

Product Name	Cat. No.	PKG size	Price
His-Detect In-Gel Stain	08500-94	100 ml	\$ 50.00
	08500-65	500 ml	\$ 189.00

### TurboTEV Protease & Turbo3C Protease

#### **Features**

- · Activity over a broad temperature (4°C to 37°C) and pH (6.5 to 8.5) range
- Both GST and His tags to facilitate its removal from the digested protein sample

### **Ordering Information**

Product Name	Cat. No.	PKG size	Price
	NU0102S	1,000 units (0.1 mg)	\$ 65.00
TurboTEV (TEV Protease) 2 mg/ml	NU0102M	10,000 units (1 mg)	\$ 466.00
	NU0102L	100,000 units (10 mg)	\$ 3,960.00
urbo3C (HRV3C Protease) 2 mg/ml	NU0101S	1,000 units (1 mg)	\$ 327.00
	NU0101M	10,000 units (10 mg)	\$ 2,723.00

His-Detect In-Gel Stain



TurboTEV (TEV Protease) Turbo3C (HRV3C Protease)

# **Cell Lysis Solution**

### **RIPA Buffer**

RIPA Buffer (10x) is a ready-to-use solution containing a variety of surfactants and protease inhibitors (Cat. No.: 08714-04). Proteins lysed with RIPA Buffer can be used in western blotting, ELISA or immunoprecipitation testing regimes.

#### **Features**

- · Contains protease inhibitors (Cat. No.: 07814-04)
- Unmixes SDS solution for immunoprecipitation (Cat. No.: 07814-04)
- · Applicable BCA protein assay without buffer exchange

Product Name	RIPA Buffer (1x, ready-to-use)	RIPA Buffer (10x)		
Protease Inhibitor Cocktail	without	with		
Composition	- 50mmol/l Tris-HCl buffer (pH7.6) - 150mmol/l NaCl - 1% Nonidet® P40 - 0.5% Sodium Deoxycholate - 0.1% SDS	【1x solution with SDS】 - 50mmol/I Tris-HCI buffer (pH7.6) - 150mmol/I NaCI - 1% Nonidet® P40 - 0.5% Sodium Deoxycholate - Protease Inhibitor Cocktail (1x) - (0.1% SDS)		
Volume/Content	100 mL	- RIPA Buffer(10x): 2ml×5本 - SDS Solution(1% SDS): 2ml×5本		

### Ordering Information

Product Name	Cat. No.	PKG size	Price
RIPA Buffer (10X)	08714-04	1 set	\$ 136.00
RIPA Buffer	16488-34	100 ml	\$ 97.00

# **Inhibitor Cocktails**

Product Name	Cat. No.	PKG size	Price
Protease Inhibitor Cocktail Tablets, EDTA-Free	NU01069	20 tablets	\$ 325.00
Protease Inhibitor Cocktail for Use with Mammalian Cell and Tissue Extracts (100x) DMSO Solution	25955-24	1 ml	\$ 63.00
	25955-11	5 x 1 ml	\$ 208.00
Phosphatase Inhibitor Cocktail	07574-61	1 ml	\$ 136.00
Phosphatase Inhibitor Cocktail (EDTA free) (100X)	07575-51	1 ml	\$ 136.00





Protease Inhibitor Cocktail. Tablets



Protease Inhibitor Cocktail



Phosphatase Inhibitor Cocktail)



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www.nacalaiusa.com





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