



## PRODUCT INFORMATION

# Thermo Scientific PageRuler Unstained Protein Ladder

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**Assembling Lot 00000000**

**Filling Lot 00000000**

**Expiry Date MM.YYYY**

**Store at -20°C**

Components	#26614	#26615
PageRuler Unstained Protein Ladder	2 x 250 µL	25 µL

[www.thermofisher.com](http://www.thermofisher.com)

[www.thermoscientific.com/pierce](http://www.thermoscientific.com/pierce)

**For Research Use Only.** Not for use in diagnostic procedures.

## Introduction

The Thermo Scientific™ PageRuler™ Unstained Protein Ladder consists of a unstained mixture of 14 recombinant, highly purified proteins ranging from 10 kDa to 200 kDa. The ladder is visualized by SDS-PAGE using coomassie or silver stains or detected in Western blots with protein stains. For easy reference, the 50 kDa protein band has a greater intensity than the other proteins in the ladder. The protein ladder is conveniently packaged and ready to use with no heating, diluting or additional reducing agent necessary.

**Storage Buffer:** 62.5 mM Tris•H<sub>3</sub>PO<sub>4</sub> (pH 7.5 at 25 °C), 1 mM EDTA, 2 % (w/v) SDS, 100 mM DTT, 1 mM NaN<sub>3</sub>, 0.01 % bromophenol blue and 33 % (v/v) glycerol.

## Important Product Information

- Do not boil the protein ladder.
- In high-percentage gels (14-18 %), large proteins (150-200 kDa) may not separate.
- In low-percentage gels (4-8 %), low-molecular weight proteins may migrate with the dye front.
- The large proteins (> 100 kDa) in the ladder may require longer transfer times or higher transfer voltages for Western blotting.
- If additional bands appear in the protein ladder, add newly prepared dithiothreitol (DTT) solution to 100 mM final concentration. DTT oxidation in the storage buffer can cause the appearance of additional bands.

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## Procedure for Using the Protein Ladder in Polyacrylamide Gel Electrophoresis

1. Thaw the ladder at room temperature. Do not boil protein ladder.
2. Mix the solution gently and thoroughly to ensure it is homogeneous.
3. Load an appropriate volume of the ladder on the gel.
  - Mini-gel: 5  $\mu\text{L}$  per well (0.75-1.0 mm thick) or 10  $\mu\text{L}$  per well (1.5 mm thick)
  - Large gel: 10  $\mu\text{L}$  per well (0.75-1.0 mm thick) or 20  $\mu\text{L}$  per well (1.5 mm thick)

**Note:** Dilute the ladder approximately 1/10 in reducing sample buffer for silver staining.

4. Return the unused protein ladder to  $-20\text{ }^{\circ}\text{C}$ .

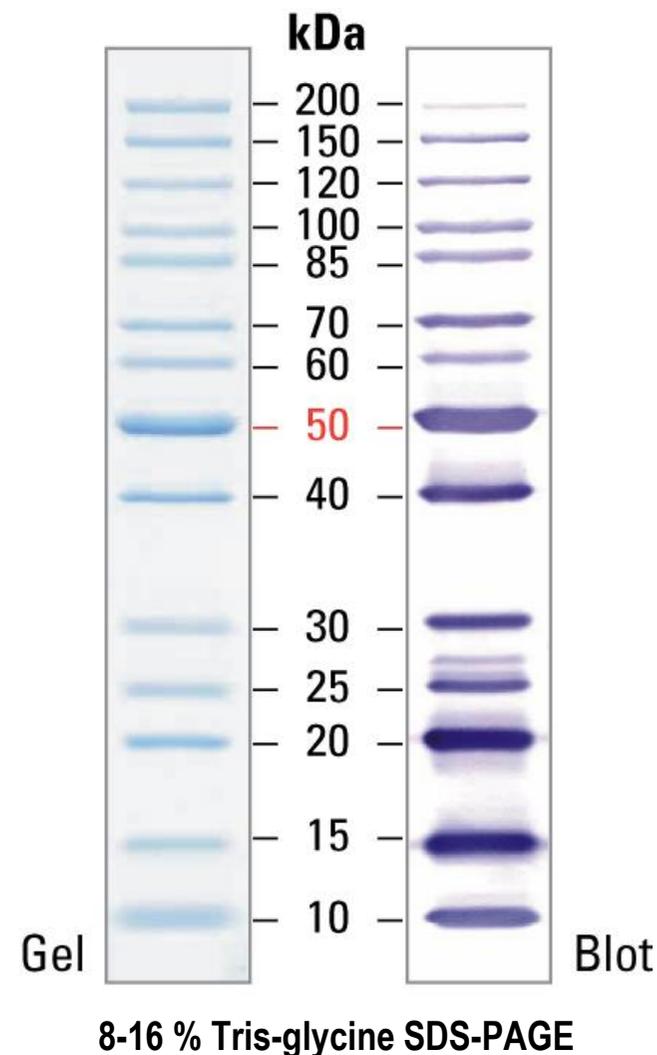
### Note

To avoid overloading gels which will be subsequently silver stained, dilute the ladder in protein loading buffer just prior to use:

Water, nuclease-free (#R0581)	To final 50 $\mu\text{L}$ volume
4X Protein Loading Buffer (e.g. LDS Sample Buffer, Non-Reducing #R84788, <a href="http://www.thermoscientific.com/pierce">www.thermoscientific.com/pierce</a> )	12.5 $\mu\text{L}$
Reducing Agent	to 50mM final conc.
Protein ladder	1 $\mu\text{L}$

Load 5  $\mu\text{L}$  of the diluted ladder per well for a mini gel/blot and 10  $\mu\text{L}$  per well for a large gel/blot.

## PageRuler Unstained Protein Ladder



## General References

- Burnette, W.N. (1981). "Western blotting": electrophoretic transfer of proteins from sodium dodecyl sulfate – polyacrylamide gels to unmodified nitrocellulose and radiographic detection with antibody and radioiodinated protein A. *Anal Biochem* 112(2):195-203.
- Kurien, B.T. and Scofield, R.H. (2003). Protein blotting: a review. *J Imm Meth* 274:1-15.
- Laemmli, U.K. (1970). Cleavage of structural proteins during the assembly of the head of bacteriophage T4. *Nature* 227:680-5.
- Towbin, H., et al. (1979). Electrophoretic transfer of proteins from polyacrylamide gels to nitrocellulose sheets: procedure and some applications. *Proc Natl Acad Sci USA* 76:4350-4.

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