Ordering Information

Catalog Number	Product Description
Molecular Weight Standards	
161-0303	**SDS-PAGE Standards,** High, 200 µl
161-0304	**SDS-PAGE Standards,** Low, 200 µl
161-0317	**SDS-PAGE Standards,** Broad, 200 µl
161-0314	**Silver Stain SDS-PAGE Standards,** Low, 200 µl
161-0315	**Silver Stain SDS-PAGE Standards,** High, 200 µl
161-0306	**Biotinylated SDS-PAGE Standards,** Low, 250 µl
161-0311	**Biotinylated SDS-PAGE Standards,** High, 250 µl
161-0319	**Biotinylated SDS-PAGE Standards,** Broad, 250 µl
161-0320	**2-D SDS-PAGE Standards**
161-0326	**Polypeptide SDS-PAGE Standards,** 200 µl
Prestained Standards	
161-0305	**Prestained SDS-PAGE Standards,** Low, 500 µl
161-0309	**Prestained SDS-PAGE Standards,** High, 500 µl
161-0318	**Prestained SDS-PAGE Standards,** Broad, 500 µl
161-0324	**Kaleidoscope Prestained Standards,** 500 µl
161-0325	**Kaleidoscope Polypeptide Standards,** 500 µl
IEF Standards	
161-0310	**IEF Standards,** pI range 4.45–9.6, 250 µl

Bio-Rad Laboratories, 2000 Alfred Nobel Dr., Hercules, CA 94547

4006048 Rev D

Silver Stain SDS-PAGE Standards, High Range

Catalog Numbers 161-0315

Product shipped on dry ice.
Store at -20 °C upon arrival.

BIO-RAD

SDS-PAGE Molecular Weight Standards, High Range Specifications

	High Range
Range	45,000 to 200,000 daltons
Contents	Rabbit skeletal muscle myosin *E. coli* β-galactosidase Rabbit muscle phosphorylase b Bovine serum albumin Hen egg white ovalbumin
Volume	200 µl concentrated solution
Storage	-20 °C
Shelf Life	1 year at -20 °C
Applications per vial	400 full size gels 800 mini gels
Recommended gel percentages	7.5%

Silver Stain SDS-PAGE Standards contain approximately 700 µg total protein in 50% glycerol (w/v), 300 mM NaN_3, 20 mM Tris, 4 mM EDTA. The proteins have been blended to give bands of equal intensity on SDS polyacrylamide gel systems run according to Laemmli[1] and stained with Bio-Rad Silver Stain or Silver Stain Plus. Different results may be obtained when alternative silver staining chemistries are used.

Reference

1. Laemmli, U. K., *Nature*, **227,** 680 (1970).
2. Hames, B. D. and Rickwood, D., **Gel Electrophoresis of Proteins: A Practical Approach,** Second Edition, p. 17, Oxford University Press, New York (1990).

Protocol

Dilute standards 1:20 in SDS Reducing Sample Buffer.* Heat for 5 minutes at 95 °C. Cool and load 10 µl/well for full length gels (16-20 cm) or 5 µl/well for mini gels. These load volumes and dilutions have been optimized for development with Bio-Rad Silver Stain or Silver Stain Plus for approximately 10 minutes. If silver stain development times vary, the loading volume or dilution of the standards may need to be adjusted to optimize the band intensity.

* SDS Reducing Sample Buffer (Prepare immediately before use)

ß-mercaptoethanol	25 µl
Stock sample buffer	475 µl
	500 µl

Stock Sample Buffer (Store at room temperature)

Distilled water	4.8 ml
0.5M Tris-HCl, pH 6.8	1.2 ml
Glycerol	1.0 ml
10% (w/v) SDS	2.0 ml
0.1% (w/v) bromophenol blue	0.5 ml
	9.5 ml

Use of stock sample buffer with insufficient or old ß-mercaptoethanol may result in doublets at the ovalbumin band.

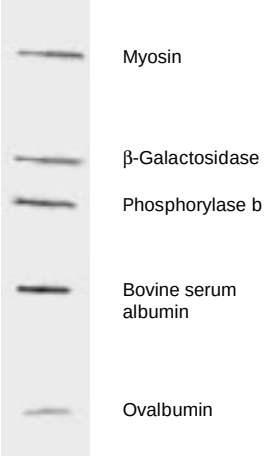

Fig. 1. Silver Stain SDS-PAGE Standards give bands of even intensities when stained with Bio-Rad's Silver Stain or Silver Stain Plus. Note that no extraneous bands are present. High range silver stain standards run on a 7.5% gel and stained with Bio-Rad's Silver Stain Kit.

Protein Molecular Weights

Protein	Molecular Weight	References
Myosin	200,000	Woods, E. F., Himmelfarb, S. and Harrington, W. F., *J. Biol. Chem.*, **238,** 2374 (1963).
***E. coli* β-galactoside**	116,250	Fowler, A. V. and Zabin, I., *Proc. Natl. Acad. Sci. USA*, **74,** 1507 (1977).
Rabbit muscle phosphorylase b	97,400	Titani, K., et. al., *Proc. Natl. Acad. Sci. USA*, **74,** 11, 4762 (1977).
Bovine serum albumin (BSA)	66,200	Brown, J. R., *Fed. Proc.*, **34,** 591 (1975).
Hen egg white ovalbumin	45,000	Warner, R. C., "Egg Proteins," in: **The Proteins,** Vol. IIA, p. 435 (Neurath, H. and Bailey, K., eds.), Academic Press, New York (1954).

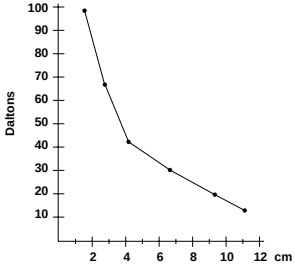

Fig. 2. Curve generated by plotting the molecular weight of the low range molecular weight standards run on a 12% SDS polyacrylamide gel vs. the distance migrated from the interface of the stacking and separating gels in centimeters. An alternative method is to plot the $\log_{10}$ relative mobility (R_f) vs. the gel concentration, %T, (percentage total monomer, i.e. grams acrylamide plus bis acrylamide/100ml).

$$R_f = \frac{\text{distance migrated by protein}}{\text{distance migrated by dye}}$$

The curve can be used to determine molecular weights of unknown proteins.[2]