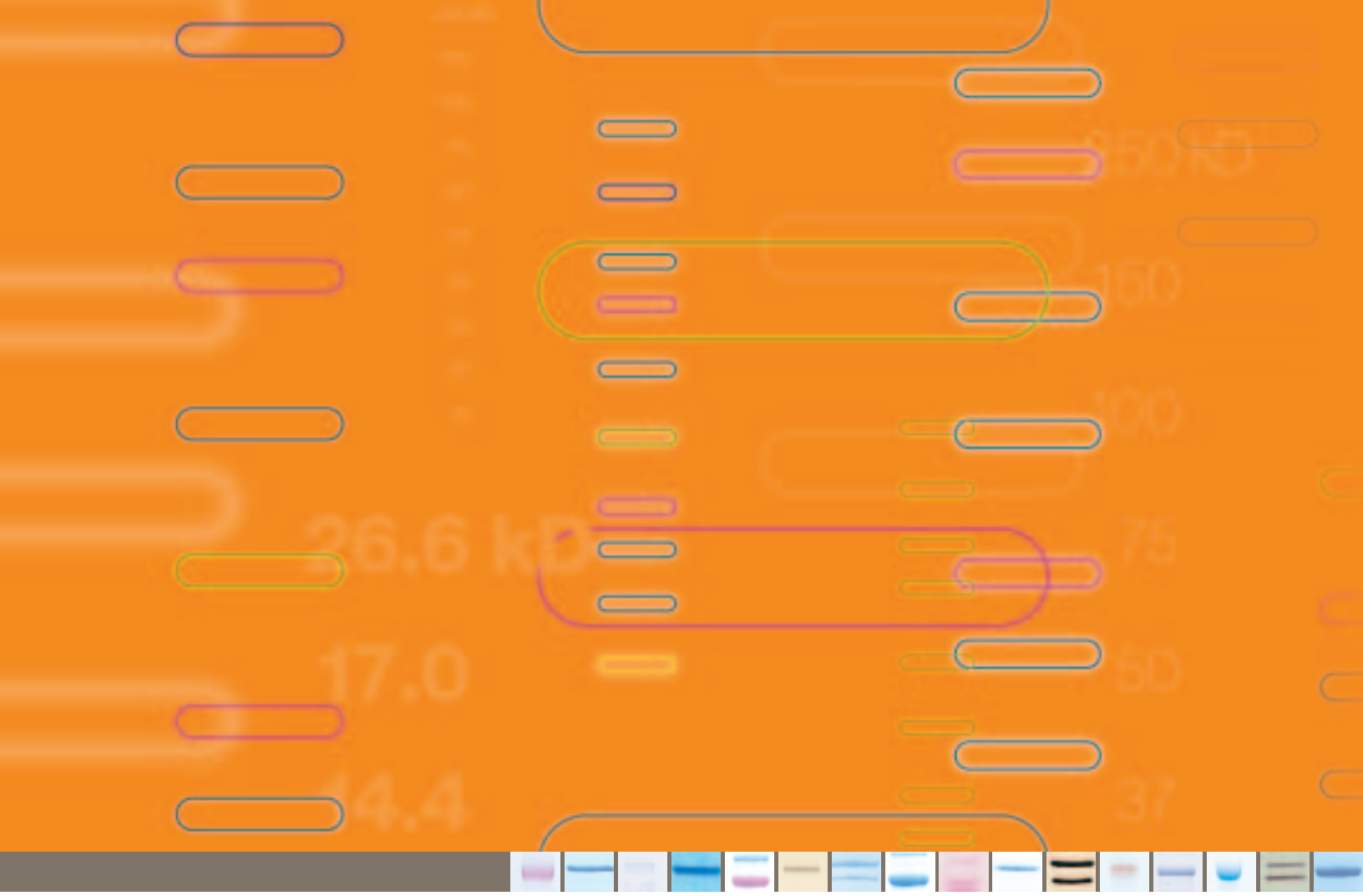


Standards for
Electrophoresis and Blotting

A Wide Range for All Applications

BIO-RAD

A Wide Range of Standards

Bio-Rad's standards provide an excellent means of monitoring electrophoresis and blotting experiments. Bio-Rad has formulated and blended a variety of protein standards for a large range of applications, including SDS-PAGE, western blotting, 2-D PAGE, and isoelectric focusing (IEF). These standards can be used for gel and blot orientation, to monitor transfer efficiency, and for molecular weight (MW) estimation and determination.

The variety of standards available from Bio-Rad can be divided into the following general categories:

SDS-PAGE and Western Blotting Standards

- Recombinant prestained protein standards
- Recombinant unstained protein standards
- Natural prestained SDS-PAGE protein standards
- Natural unstained SDS-PAGE protein standards

Specialty Standards

- Protein standards for IEF
- Protein standards for 2-D SDS-PAGE

Bio-Rad's protein standard selection guide.

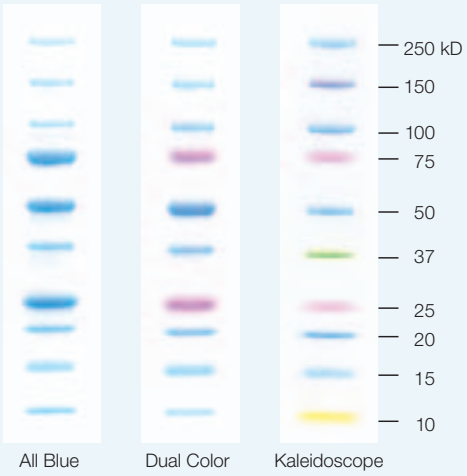
	Recombinant	Natural							
Application	Precision Plus Protein™ all blue, dual color, Kaleidoscope™, and unstained	Prestained SDS-PAGE high, low, and broad range	Kaleidoscope prestained and polypeptide	SDS-PAGE high, low, and broad range	Biotinylated SDS-PAGE high, low, and broad range	Silver stain SDS-PAGE high and low range	IEF	2-D SDS-PAGE	Unstained polypeptide
Electrophoresis									
SDS-PAGE	●	●	●	●	●	●	○	○	○
Peptide electrophoresis	○	○	●*	○	○	○	○	○	●
IEF	○	○	○	○	○	○	●	○	○
2-D electrophoresis	○	○	○	○	○	○	●	●	○
MW determination	●	○	○	●	●	●	○	○	●
MW estimation	●	◒	◒	●	●	●	○	○	●
Silver staining	◒**	○	○	◒	○	●	○	○	◒
Blotting									
Transfer efficiency	●***	●	●	○	○	○	○	○	○
Immunodetection compatibility	●**	○	○	○	●	○	○	○	○
Catalog numbers, in order listed above	161-0373 161-0374 161-0375 161-0363	161-0309 161-0305 161-0318	161-0324 161-0325	161-0303 161-0304 161-0317	161-0311 161-0306 161-0319	161-0315 161-0314	161-0310	161-0320	161-0326

● = Good
◒ = Acceptable
○ = Not applicable

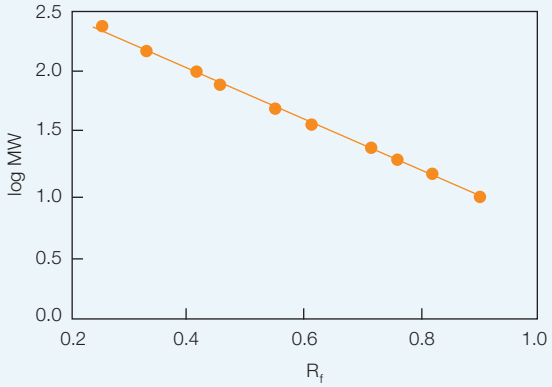
* Polypeptide standards only
** Unstained standards only
*** All blue, dual color, and Kaleidoscope standards only

Bio-Rad offers a complete family of Precision Plus Protein prestained standards, including all blue, dual color, and Kaleidoscope options. All standards show the same pattern, with only minimal shift, and can be used for MW determination of unknown proteins in gels and on blots. Precision Plus Protein prestained standards offer:

- Exceptional linearity ($r^2 > 0.99$) for determining MW
- 10 sharp, nonshifting bands (10–250 kD)
- Lot-to-lot consistency
- Matching migration patterns among the entire Precision Plus Protein standards family
- 3 high-intensity reference bands in the all blue and dual color standards — at 25, 50, and 75 kD



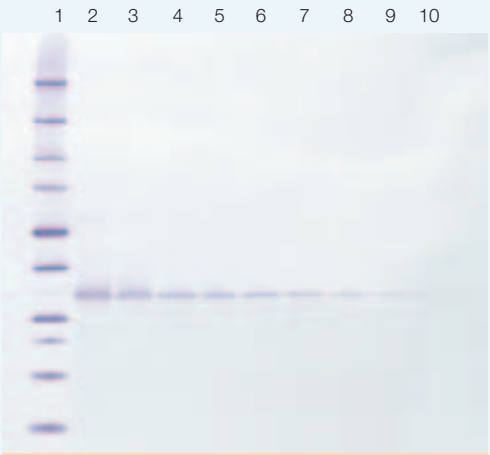
Precision Plus Protein prestained standards family.



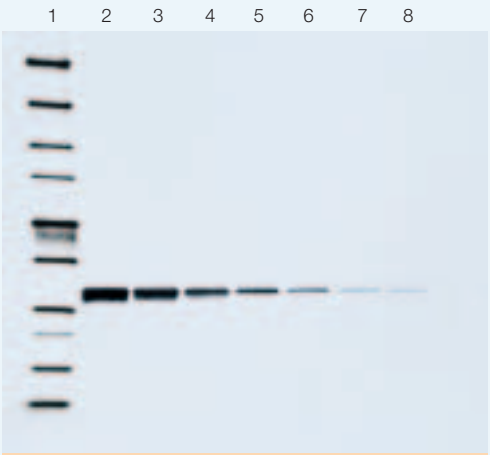
Exceptional linearity of Precision Plus Protein standards. The standard curve was generated by plotting log MW vs. migration distance (R_f) of each protein standard band through an SDS-PAGE gel. Precision Plus Protein Kaleidoscope standards showed $r^2 = 0.996$, demonstrating a strong linear relationship between the proteins' log MW and migration distance on a gel. All Precision Plus Protein prestained and unstained standards generate standard curves with r^2 values ≥ 0.99.

Recombinant unstained protein standards allow accurate MW determination on SDS-PAGE gels and western blots. Precision Plus Protein unstained standards have the following attributes:

- The bands in every batch have the same MW, confirmed by mass spectrometry and migration in a Laemmli SDS-PAGE buffer system
- Unique *Strep*-tag affinity sequence allows detection and MW determination on western blots
- 10 sharp, nonshifting bands (10–250 kD)
- 3 high-intensity reference bands — at 25, 50, and 75 kD



Western blot detection of Green Fluorescent Protein (GFP) and Precision Plus Protein unstained standards using the Immun-Blot® alkaline phosphatase (AP) colorimetric detection kit. Maximum sensitivity achievable with Immun-Blot AP is 100 pg. A gel run with 4 µl of standards (lane 1) and a dilution series of *E. coli* lysate containing overexpressed GFP (lanes 2–10) was transferred to a PVDF membrane. The blot was probed with a primary antibody specific for GFP, then incubated with StrepTactin-AP and a secondary antibody conjugated to AP. The blot was developed using the Immun-Blot AP kit.



Western blot detection of GFP and Precision Plus Protein unstained standards using the Immun-Star™ horseradish peroxidase (HRP) chemiluminescent detection kit. Maximum sensitivity achievable with Immun-Star HRP is 1–3 pg. A gel run with 0.5 µl of standards (lane 1) and a dilution series of *E. coli* lysate containing overexpressed GFP (lanes 2–8) was transferred to a PVDF membrane. The blot was probed with a primary antibody specific for GFP, then incubated with StrepTactin-HRP and a secondary antibody conjugated to HRP. The blot was developed using the Immun-Star HRP kit.

Natural Prestained SDS-PAGE Protein Standards

Prestained standards are visualized before the gel is stained, making them ideal for monitoring protein migration during an electrophoretic run, for gel and blot orientation, and for assessing transfer efficiency.

Prestained SDS-PAGE Standards

- Available in high, low, and broad ranges
- Blended proteins give uniform band intensities
- Covalently bound dye will not dissociate during normal staining or destaining

Kaleidoscope Prestained and Polypeptide Standards

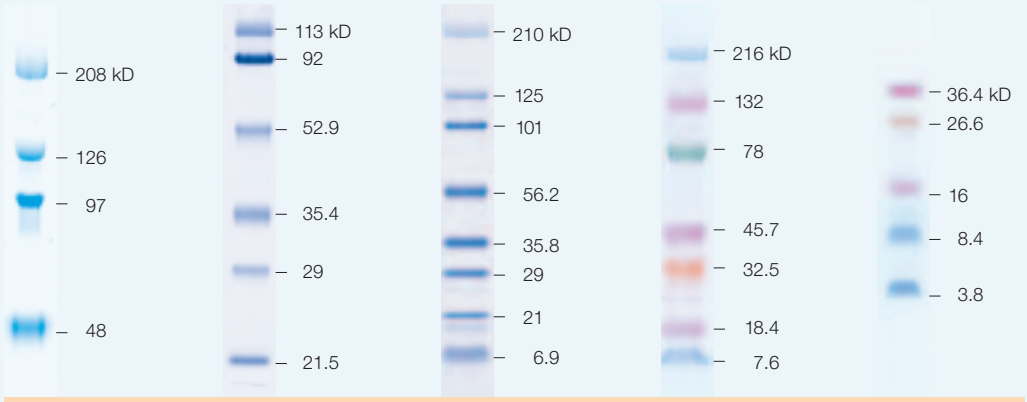
These standards include all attributes of the prestained SDS-PAGE standards, plus:

- Individually colored proteins for instant band recognition
- Good transfer efficiency for western blotting
- Specially formulated polypeptide standards for resolving peptides on Tris-Tricine gels

Constituent proteins of natural prestained SDS-PAGE standards.

			Prestained SDS-PAGE Standards			Kaleidoscope Standards	
Protein	Source	Approximate MW* (kD)	High	Low	Broad	Prestained	Polypeptide
Myosin	Rabbit skeletal muscle	200.0	●		●	●	
-Galactosidase	*E. coli*	116.3	●		●	●	
Phosphorylase b	Rabbit muscle	97.4		●			
Serum albumin (BSA)	Bovine	66.2	●	●	●	●	
Ovalbumin	Hen egg white	45.0	●	●	●		
Carbonic anhydrase	Bovine	31.0		●	●	●	●
Trypsin inhibitor	Soybean	21.5		●	●	●	●
Lysozyme	Hen egg white	14.4		●	●	●	●
Aprotinin	Bovine pancreas	6.5			●	●	●
Insulin, B chain, oxidized	Bovine	3.5					●

* MW will vary from lot to lot; see lot-specific calibration included with standards.



Natural prestained standards. Molecular weights shown are of representative lots. Actual weights may vary.

Natural Unstained SDS-PAGE Protein Standards

Natural unstained protein standards allow accurate MW determination on SDS-PAGE gels. Every batch is tested for proper mobility, providing a reliable control for gel-to-gel variability.

SDS-PAGE Standards

- Available in high, low, and broad MW ranges
- Blended to give uniform band intensities with Coomassie Blue R-250 stain

Polypeptide SDS-PAGE Standards

- Formulated for MW determination of peptides and small proteins resolved on Tris-Tricine gels
- Contain 6 polypeptides with MW ranging from 1.4 to 26.6 kD
- Blended to stain uniformly with Coomassie G-250 stain

Silver Stain SDS-PAGE Standards

- Blended to give uniform band intensities with high-sensitivity silver stain kits
- Available in high and low MW ranges

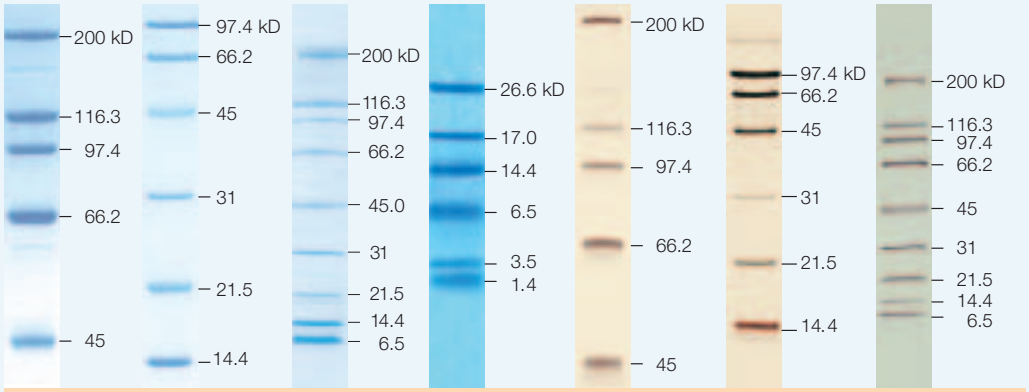
Biotinylated SDS-PAGE Standards

- Contain proteins conjugated to biotin, which can be detected with either avidin or streptavidin conjugates
- Available in high, low, and broad MW ranges

Constituent proteins of natural unstained SDS-PAGE standards.

			Ranges Available*			
Protein	Source	MW (kD)	High	Low	Broad	Polypeptide
Myosin	Rabbit skeletal muscle	200.0	●		●	
β-Galactosidase	*E. coli*	116.3	●		●	
Phosphorylase b	Rabbit muscle	97.4	●	●	●	
Serum albumin	Bovine	66.2	●	●	●	
Ovalbumin	Hen egg white	45.0	●	●	●	
Carbonic anhydrase	Bovine	31.0		●	●	
Triosephosphate isomerase	Rabbit	26.6				●
Trypsin inhibitor	Soybean	21.5		●	●	
Myoglobin	Equine	17.0				●
α-Lactalbumin	Bovine	14.5				●
Lysozyme	Hen egg white	14.4		●	●	
Aprotinin	Bovine pancreas	6.5			●	●
Insulin B chain, oxidized	Bovine	3.5				●
Bacitracin	*Bacillus licheniformis*	1.4				●

* SDS-PAGE — high, low, broad, and polypeptide; silver stain — high and low; biotinylated — high, low, and broad.



SDS-PAGE Standards, High Range | SDS-PAGE Standards, Low Range | SDS-PAGE Standards, Broad Range | Polypeptide SDS-PAGE Standards | Silver Stain SDS-PAGE Standards, High Range | Silver Stain SDS-PAGE Standards, Low Range | Biotinylated SDS-PAGE Standards, Broad Range

Natural unstained standards.

Specialty Protein Standards for IEF

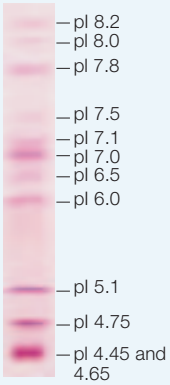
IEF Standards

- Allow reproducible, dependable pI calibration in native polyacrylamide and agarose IEF gels
- Contain 9 native proteins with pI ranging from 4.45 to 9.6
- 5 of the 9 proteins are naturally colored to monitor focusing

Constituent proteins of IEF standards.*

Protein	Color	pI
Cytochrome c	Red	9.6
Lentil lectin (3 bands)	—	7.8, 8.0, 8.2
Human hemoglobin C	Red	7.5
Human hemoglobin A	Red	7.1
Equine myoglobin (2 bands)	Brown	7.0
Human carbonic anhydrase	—	6.5
Bovine carbonic anhydrase	—	6.0
□-Lactoglobulin B	—	5.1
Phycocyanin (3 bands)	Blue	4.45, 4.65, 4.75

* Because the IEF standards are in native form, they cannot be used with reducing or denaturing agents such as urea, □-mercaptoethanol, or dithiothreitol. For calibration of IEF tube gels containing urea, use 2-D SDS-PAGE standards.



IEF standards. The gel was stained with Crocein Scarlet.

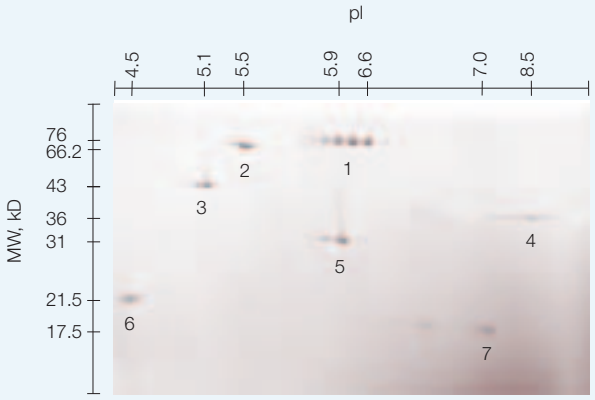
Specialty Protein Standards for 2-D SDS-PAGE

2-D SDS-PAGE Standards

- Provide calibrated references for the pI and MW of proteins in 2-D SDS-PAGE applications
- Consist of 7 reduced, denatured proteins that can be visualized with silver or Coomassie Blue staining

Constituent proteins of 2-D SDS-PAGE standards.

Protein	pI	MW (kD)
1. Hen egg white conalbumin	6.0, 6.3, 6.6	76
2. Bovine serum albumin (BSA)	5.4, 5.5, 5.6 (empirically determined)	66.2
3. Bovine muscle actin	5.0, 5.1 (empirically determined)	43
4. Rabbit muscle GAPDH	8.3, 8.5	36
5. Bovine carbonic anhydrase	5.9, 6.0	31
6. Soybean trypsin inhibitor	4.5	21.5
7. Equine myoglobin	7.0	17.5



Two-dimensional electrophoretic protein pattern of 2-D SDS-PAGE standards. The standards (2.5 µl) were run on 7 cm ReadyStrip™ IPG strips, then in the Mini-PROTEAN® II cell. For method details, see Klose (1975), Klose and Feller (1981), and Jungblut and Seifert (1990). The gel was stained with Bio-Rad's silver stain kit.

References

Jungblut PR and Seifert R, Analysis by high-resolution two-dimensional electrophoresis of differentiation-dependent alterations in cytosolic protein pattern of HL-60 leukemic cells, J Biochem Biophys Method 21, 47–58 (1990)

Klose J, Protein mapping by combined isoelectric focusing and electrophoresis of mouse tissues. A novel approach to testing for induced point mutations in mammals, Humangenetik 26, 231–243 (1975)

Klose J and Feller M, Two-dimensional electrophoresis of membrane and cytosol proteins of mouse liver and brain, Electrophoresis 2, 12–24 (1981)

Ordering Information

Catalog #	Description
Recombinant Prestained Protein Standards	
161-0373	Precision Plus Protein All Blue Standards, 500 µl
161-0374	Precision Plus Protein Dual Color Standards, 500 µl
161-0375	Precision Plus Protein Kaleidoscope Standards, 500 µl
Recombinant Unstained Protein Standards	
161-0363	Precision Plus Protein Unstained Standards, 1 ml
Natural Prestained SDS-PAGE Protein Standards	
161-0324	Kaleidoscope Prestained Standards, 500 µl
161-0325	Kaleidoscope Polypeptide Standards, 500 µl
161-0305	Prestained SDS-PAGE Standards, low range, 500 µl
161-0309	Prestained SDS-PAGE Standards, high range, 500 µl
161-0318	Prestained SDS-PAGE Standards, broad range, 500 µl
Natural Unstained SDS-PAGE Protein Standards	
161-0303	SDS-PAGE Standards, high range, 200 µl
161-0304	SDS-PAGE Standards, low range, 200 µl
161-0317	SDS-PAGE Standards, broad range, 200 µl
161-0326	Polypeptide SDS-PAGE Standards, 200 µl
161-0315	Silver Stain SDS-PAGE Standards, high range, 200 µl
161-0314	Silver Stain SDS-PAGE Standards, low range, 200 µl
161-0311	Biotinylated SDS-PAGE Standards, high range, 250 µl
161-0306	Biotinylated SDS-PAGE Standards, low range, 250 µl
161-0319	Biotinylated SDS-PAGE Standards, broad range, 250 µl
Specialty Protein Standards	
161-0310	IEF Standards, 250 µl
161-0320	2-D Standards, 250 µl
Biotinylated SDS-PAGE Standards Kits	
161-0312	Biotinylated SDS-PAGE Standards Kit, high range, avidin-HRP
161-0313	Biotinylated SDS-PAGE Standards Kit, high range, avidin-AP
161-0307	Biotinylated SDS-PAGE Standards Kit, low range, avidin-HRP
161-0308	Biotinylated SDS-PAGE Standards Kit, low range, avidin-AP
161-0321	Biotinylated SDS-PAGE Standards Kit, broad range, avidin-HRP
161-0322	Biotinylated SDS-PAGE Standards Kit, broad range, avidin-AP

Catalog #	Description
Accessory Reagents	
161-0380	Precision Protein™ StrepTactin-HRP Conjugate, 300 µl
161-0382	Precision Protein StrepTactin-AP Conjugate, 300 µl
170-6528	Avidin-HRP, 2 ml
170-6533	Avidin-AP, 1 ml
170-3554	Streptavidin-AP, 0.5 ml
Premixed Sample Buffers	
161-0737	Laemmli Sample Buffer, 30 ml
161-0738	Native Sample Buffer, 30 ml
161-0739	Tricine Sample Buffer, 30 ml
161-0768	TBE-Urea Sample Buffer, 30 ml
161-0763	IEF Sample Buffer, 30 ml
161-0764	Zymogram Sample Buffer, 30 ml
161-0767	Nucleic Acid Sample Buffer, 5x, 10 ml
161-0791	XT Sample Buffer, 4x, 10 ml
Gel-Casting Buffers	
161-0799	Stacking Gel Buffer, 0.5 M Tris-HCl, pH 6.8, 1 L
161-0798	Resolving Gel Buffer, 1.5 M Tris-HCl, pH 8.8, 1 L
Premixed Electrophoresis Buffers	
161-0732	10x Tris/Glycine/SDS, 1 L
161-0772	10x Tris/Glycine/SDS, 5 L cube
161-0734	10x Tris/Glycine, 1 L
161-0771	10x Tris/Glycine, 5 L cube
161-0744	10x Tris/Tricine/SDS, 1 L
161-0761	10x IEF Anode Buffer, 250 ml
161-0762	10x IEF Cathode Buffer, 250 ml
161-0733	10x Tris/Boric Acid/EDTA (TBE), 1 L
161-0770	10x Tris/Boric Acid/EDTA (TBE), 5 L cube
161-0741	10x Tris/Boric Acid/EDTA (TBE), extended range, 1 L
161-0743	50x Tris/Acetic Acid/EDTA (TAE), 1 L
161-0773	50x Tris/Acetic Acid/EDTA (TAE), 5 L cube
161-0765	10x Zymogram Renaturation Buffer, 125 ml
161-0766	10x Zymogram Development Buffer, 125 ml

BIO-RAD

Bio-Rad Laboratories, Inc.

Life Science Group

Web site *www.bio-rad.com* ***USA*** *(800) 4BIORAD* ***Australia*** *02 9914 2800* ***Austria*** *(01)-877 89 01* ***Belgium*** *09-385 55 11* ***Brazil*** *55 21 2527 3454* ***Canada*** *(905) 712-2771* ***China*** *(86-21) 63052255* ***Czech Republic*** *+ 420 2 41 43 05 32* ***Denmark*** *44 52 10 00* ***Finland*** *09 804 22 00* ***France*** *01 47 95 69 65* ***Germany*** *089 318 84-0* ***Hong Kong*** *852-2789-3300* ***Hungary*** *36 1 455 8800* ***India*** *(91-124)-6398112/113/114, 6450092/93* ***Israel*** *03 951 4127* ***Italy*** *39 02 216091* ***Japan*** *03-5811-6270* ***Korea*** *82-2-3473-4460* ***Latin America*** *305-894-5950* ***Mexico*** *55-52-00-05-20* ***The Netherlands*** *0318-540666* ***New Zealand*** *64 9 415 2280* ***Norway*** *23 38 41 30* ***Poland*** *+ 48 22 331 99 99* ***Portugal*** *351-21-472-7700* ***Russia*** *7 095 721 1404* ***Singapore*** *65-6415 3188* ***South Africa*** *00 27 11 4428508* ***Spain*** *34 91 590 5200* ***Sweden*** *08 555 12700* ***Switzerland*** *061 717-9555* ***Taiwan*** *(8862) 2578-7189/2578-7241* ***United Kingdom*** *020 8328 2000*