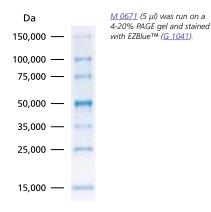
Recombinant Markers

The Recombinant Molecular Weight Standard Mixture contains seven precisely sized proteins, molecular weights 15, 25, 35, 50, 75, 100, and 150 kDa. Recombinant proteins form sharp bands for accurate sample MW calculations.

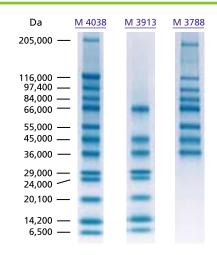
Product Code	Description	MW Range (Da)
M 0671	Recombinant Molecular Weight	15,000-150,000
	Standard Mixture	



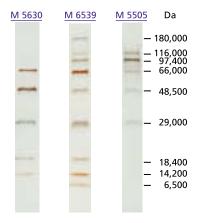
SigmaMarker™

SigmaMarkers encompass the range of molecular weights common to most proteins and their subunits. Lyophilized with sample buffer, the wide, high, and low range markers are ready for use following reconstitution with water. They are formulated to yield a distribution of well-defined bands of approximately equal intensity following electrophoresis and Coomassie® Blue staining.

Product Code	Description	MW Range (Da)
M 4038	SigmaMarker	Wide (6,500-205,000)
M 3913	SigmaMarker	Low (6,500-66,000)
M 3788	SigmaMarker	High (36,000-205,000)



 \underline{M} 4038, \underline{M} 3913, and \underline{M} 3788 (5 μ l) were run on a 4-20% PAGE gel and stained with EZBlue (\underline{G} 1041).



 \underline{M} 5630, \underline{M} 6539, and \underline{M} 5505 (7.5 μ l of a 1:15 dilution) were run on a 10-18% SDS-PAGE gel and

M 3546 (5 μl of a 1:20 dilution) was run on a 10-20%

Tris-Tricine SDS-PAGE gel, fixed

with 5% glutaraldehyde and

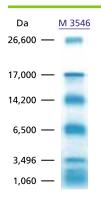
stained with EZBlue™

(G 1041).

Silver Stain Markers

Designed for molecular weight determinations on silver stained gels, Silver Stain SDS-PAGE molecular weight standard mixtures contain evenly distributed, well resolved proteins of equal intensity. See also ProteoSilver™, page 110.

Product Code	Description	MW Range (Da)
<u>M 5630</u>	Silver Stain SDS-PAGE Molecular Weight Standard	Low 14,000-66,000
M 6539	Silver Stain SDS-PAGE Molecular Weight Standard	Wide 6,500-180,000
<u>M 5505</u>	Silver Stain SDS-PAGE Molecular Weight Standard	High 29,000-116,000



Ultra-Low Range Markers

Sigma's Ultra-Low Range molecular weight marker is recommended for use with Tris-Tricine SDS-PAGE systems. Glutaraldehyde fixing is suggested to retain low molecular weight proteins.

Product Code	Description	MW Range (Da)
M 3546	Molecular Weight Marker for SDS-PAGE	Ultra-Low 1,000-26,600

Protein Staining Reagents

To meet the great diversity of protein analysis needs, Sigma offers a wide selection of protein visualization (staining) reagents. EZBlueTM and ProteoSilverTM, designed specifically for proteomics, also perform impressively in traditional PAGE formats.

Protein Dyes and Stains Selection Chart

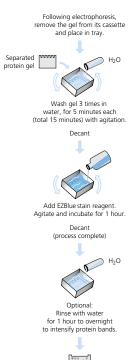
																									$\overline{}$
Optimize your protein detection by choosing the best reagent for your application. ✓ Good Choice ★ BEST Choice		Alcian Blue 8GX — $ ext{A} ext{3157}$	Amido Black Staining Solution — $\overline{ ext{A}8181}$	Brilliant Blue G Concentrate — <u>B 8522</u>	Brilliant Blue R Staining Solution — $\overline{ ext{B} ext{ } 6529}$	Coomassie TM Violet R 200 — $\overline{27817}$	Eosin Y — <u>45235</u>	EZBlue™ — <u>G 1041</u>	Fast Green FCF — <u>F 7252</u>	Fixing Solution — <u>F 7264</u>	Fluorescamine — <u>F 9015</u>	GlycoProfile III — PP0300	GlycoProtein Detection Kit (Colorimetric) — <u>GLYCO-PRO</u>	Gold Solution, Colloidal — $\overline{50755}$	Oil Red O — <u>O 9755</u>	Ponceau S Solution — P 7170	ProteoSilver TM — PROT-SIL1	ProteoSilver Plus — <u>PROT-SIL2</u>	Reversible Protein Detection Kit — R-PROB	Sudan Black — <u>86015</u>	SYPRO® Orange Protein Gel Stain — <u>§ 5692</u>	SYPRO® Red Protein Gel Stain — <u>S 5817</u>	SYPRO® Ruby Protein Blot Stain — <u>S 4817</u>	SYPRO® Ruby Protein Gel Stain — S 4942	SYPRO® Tangerine Protein Gel Stain — <u>5 5942</u>
Colorimetric		V	V	V	V	V	V	*	/				V	V	V	V	V	V	1	/					
Colorimetric Fluorescent											V	1									*	*	*	*	*
	Proteins		V	V	/	V	/	*	1		V			/		/	*	*	1		/	1	1	1	1
on	Glycoproteins											*	*											1	
ecti	Glycoproteins Lipids and/or Lipoproteins Nucleic Acids														*					V				1	
Det	Nucleic Acids																/	V							
	Phospholipids, Neutral Fats, & Sterols																			*					
	PAGE	1	V	V	V		V	V	V	V		V	V				1	V	1	V	V	V		V	1
	IEF (Acrylamide)			1	/	*			/															1	
Agarose Gel			V		*															V					
PVDF Membrane				V	*			V				V	V			V			V				V		
Agarose Gel PVDF Membrane Cellulose Acetate Membrane Nitrocellulose Membrane															*	/				V					
Nitrocellulose Membrane		V	V											/		*			V				V		
	Nylon Membrane																		*						
	MALDI-MS			✓	✓			✓				✓					✓	*					V	✓	V
High Sensitivity (10 ng)								*																	V
res	Higher Sensitivity (1 ng)																				/	/	✓		
Highest Sensitivity (0.1 ng/mm²)																	*	*							
Special Features	Fixing Reagent			✓	✓			✓		*							✓	V			V				V
ecia	Requires No Destaining							*									✓	V			/				V
Sp	Rapid Development (5 minutes)		✓													✓									
	Reversible						. 1									. 1		. 1	4						

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Ε

ELECTROPHORESIS







SigmaMarkerTM, Wide Range (M 4038) was serially diluted and run on a Tris-Acetate 6-15% gel. Following electrophoresis, the gel was stained with Sigma EZBlue (G 1041). The lane to the far right demonstrates detection of 5 ng of BSA and 11 ng of carbonic anhydrase.

Coomassie™ Stains

EZBlue™ Gel Staining Reagent

Convenient, sensitive, and safe, EZBlue Coomassie Brilliant Blue G-250 colloidal protein stain improves protein electrophoresis results while significantly reducing staining time. Conveniently packaged, EZBlue requires no messy weigh-ups or additions of methanol or acid. As a colloidal stain, it reacts only with proteins, not the gel itself. Background staining is reduced, so protein bands can be visualized almost immediately. No destaining step is required, although a water wash may intensify bands and clarify the background. Most impressively, EZBlue is extremely sensitive, detecting as little as 5 ng of protein.

Features & Benefits

- Pre-mixed solution eliminates the time and effort required to prepare the stain
- Increased sensitivity ensures low abundance proteins can be detected (as little as 5 ng)
- Rapid reaction significantly reduces the amount of time required to stain and rinse
- Eliminates solvent waste so you save the expense of hazardous material disposal

Brilliant Blue G Concentrate

Brilliant Blue G Concentrate is a Coomassie G-250 methanol-based stain designed for protein detection in polyacrylamide and agarose gels. Because this stain contains methanol and acetic acid, gels require no fixing step prior to staining. Each bottle dilutes to 1 L with water.

Brilliant Blue R Staining Solution

Brilliant Blue R Staining Solution is designed specifically for staining protein in SDS-PAGE and agarose gels. Because this stain contains ethanol and acetic acid, gels require no fixing step prior to staining. The gel is immersed in staining solution for 30 minutes and destained with 10% acetic acid.

Coomassie™ Violet R 200

Coomassie Violet R 200 is used for rapid detection of proteins in IEF without removal of carrier ampholytes.

Product Code	Description	Size
<u>G 1041</u>	EZBlue Gel Staining Reagent	500 mL 3.8 L
<u>B 8522</u>	Brilliant Blue G Concentrate	1 Btl
<u>B 6529</u>	Brilliant Blue R Staining Solution	1 L
27817	Coomassie Violet R 200	25 g 100 g

ProteoSilver™ and ProteoSilver Plus

Conveniently packaged, and highly sensitive, ProteoSilver and ProteoSilver Plus are ideal products for any proteomics scientist. Both kits contain prepared solutions of silver staining reagents along with detailed instructions to achieve optimal results. With a detection limit of 0.1 ng/mm² of protein (BSA) and an extremely low background, ProteoSilver leads to superior detection of low abundance proteins.

ProteoSilver Plus is MALDI-MS compatible and does not contain glutaraldehyde, which crosslinks lysine residues, resulting in inaccurate spectra. This kit contains two additional reagents for destaining an excised protein spot, for those wishing to perform further characterization through tryptic digest and MALDI-MS analysis. Both kits contain sufficient reagents for staining 25 mini-gels.

Features & Benefits

- Pre-mixed and pre-weighed solutions reduce time and cost of purchasing and preparing individual components
- **High sensitivity and low background** ensure very low abundance proteins can be detected and resolved from other proteins
- MALDI-compatible. Protein spots of interest can be further characterized by mass spectrometry using ProteoSilver Plus



Serial dilutions of SigmaMarker (<u>M. 4038</u>) were loaded onto a 4-20% gel and stained with ProteoSilver (<u>PROT-SIL1</u>). Lanes 1 and 2: 5 µl diluted 1:100. Lanes 3 and 4: 2.5 µl diluted 1:1000. Lanes 5 and 6: 10 µl diluted 1:1000. Lanes 7 and 8: 5 µl diluted 1:1000. Lanes 9 and 10: 2.5 µl diluted 1:1000. The amounts of protein for each band in lanes 9 and 10 range from 0.3 - 1 ng.



4 µg of E. coli whole cell extract was separated by IEF using a ProteoGeI™ IPG strip (pH 3-10) (L2531), equilibrated using ProteoGel IPG Equilibration Buffer (L7281), and run on a Tris-Acetate 2D 8-18% gel. Following electrophoresis, the gel was stained with ProteoSilver (PROT-SIL1).



ProteoSilver



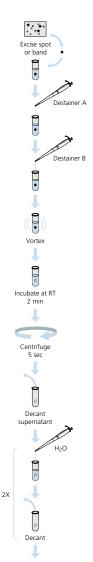
ProteoSilver Plus

Product Code	Description	Size
PROT-SIL1	ProteoSilver	1 kit
PROT-SIL2	ProteoSilver Plus	1 kit

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ELECTROPHORESIS

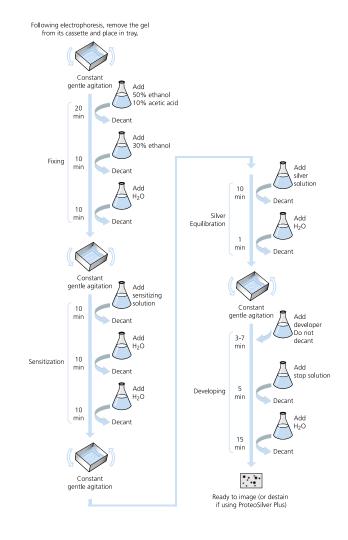
ProteoSilver Plus Destaining

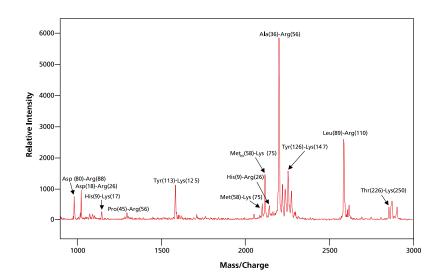


Ready for tryptic digest and MALDI-MS analysis [Proteomics Sequencing Grade Trypsin (<u>T 6567</u>)]

Bovine carbonic anhydrase (220 ng) was run on a 4-20% SDS-PAGE gel and silver stained with ProteoSilver Plus (PROT-SIL2). The silver stained band was excised from the gel and destained using ProteoSilver Plus (PROT-SIL2). The protein in the excised band was digested overnight with Proteomics Grade Trypsin (T.6567), and the peptides were extracted. The peptides were concentrated (3 μl), combined with an equal volume of α-cyano-4-hydroxycinnamic acid (C.8982) and 1 μl was spotted on the MALDI target. The peptides were analyzed in the linear positive ion mode on a Kratos® Kompact® MALDI-MS. Only the expected peptide fragments were generated. No interference was observed.

ProteoSilver™ and ProteoSilver Plus Staining





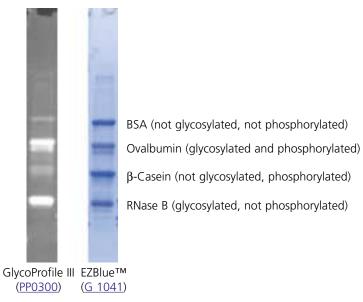


GlycoProfile™ III Fluorescent Glycoprotein Detection Kit

Identify glycoproteins with superior selectivity using Sigma's new GlycoProfile III. Glycoprotein study, or Glycomics, is a rapidly growing, dynamic new field of research. Accurate initial identification of glycoproteins plays an important role in this research. Sigma understands and meets your needs with the most selective glycoprotein detection product available.

ProteoProfile PTM Marker (Product Code <u>P 1745</u>) contains a mix of glycosylated, phosphorylated and unmodified proteins and is available individually as well as with PP0300.

*Refer to Post-Translational Modification section, p 20-24.



Superior Selectivity ProteoProfile PTM Marker (<u>P 1745</u>), containing glycosylated and non-glycosylated proteins, was separated by electrophoresis on a 4-20% SDS-PAGE gel. The gel was stained with GlycoProfile III (PP0300) (left), fluorescently imaged, and then stained for total protein with EZBlueTM Gel Staining reagent (<u>G 1041</u>) (right). Each band represents approximately 300 ng of protein.

Product Code	Description	Size
<u>PP0300</u>	GlycoProfile™ III Fluorescent Glycoprotein Detection Kit	1 kit
<u>P 1745</u>	ProteoProfile™ PTM Marker	0.1 ml

Apply ProteoProfile PTM Marker and sample(s) to PAGE gel. Separate proteins 3% acetic acid. Fix proteins in the gel 60 minutes Ultrapure 2 x 30 minutes Decant Oxidizina Oxidize carbohydrates 20 minutes Ultrapure Water Wash ge 5 minutes GlycoProfile™ III 60-90 minutes covered Ultrapure Wash gel View or image glycoproteins at Em 312 nm Optiona Stain all proteins with F7Rlue

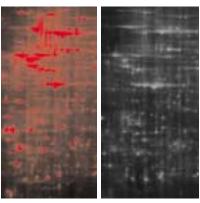
SYPRO® Ruby Gel Stain

Molecular Probes' SYPRO Ruby gel stain is a ready-to-use, ultra sensitive, luminescent stain for the detection of proteins separated by PAGE. Designed especicially for 2D PAGE, SYPRO Ruby also performs impressively in PAGE and IEF gels. Ex/Em = 280, 450/610 nm

Features & Benefits

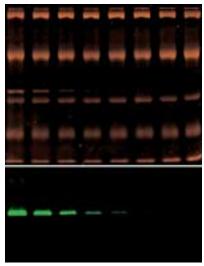
- Uses a simple staining protocol with no possibility of over-staining
- Delivers a linear quantitation range over three orders of magnitude
- Stains glycoproteins, lipoproteins, calcium binding proteins, fibrillar proteins and others
- Will not stain extraneous nucleic acids
- Sensitive to 1 ng of protein

Product Code	Description	Size
<u>S 4942</u>	SYPRO Ruby Gel Stain	1 kit

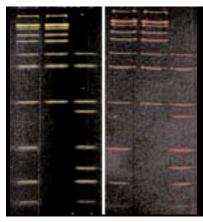


SYPRO Ruby protein gel stain compared to a silver stain. Proteins from a fibroblast cell lysate were run on identical 2-D gels and stained with SYPRO Ruby protein gel stain (left) or silver stain (right). The grayscale values of the gel stained with SYPRO Ruby dye have been inverted for easier comparison with the silver-stained gel.





Sypro Tangerine Gel stain Zymographic detection of beta-D-glucuronidase activity after visualization of total protein patterns using SYPRO Tangerine protein gel stain. Molecular weight standards containing decreasing amounts of Escherichia coli beta-glucuronidase were run on identical SDS-polyacrylamide gels and stained with SYPRO Tangerine protein gel stain (top) followed by incubation with ELF 97 D-glucuronidase substrate to detect the activity of beta-D-glucuronidase (bottom).



Protein Molecular Weight Standards separated on identical 12% SDS-PAGE gels and then stained with either SYPRO Orange (left) or SYPRO Red (right) protein gel stains.

SYPRO Tangerine Gel Stain

Molecular Probes' SYPRO Tangerine gel stain is an extremely versatile stain for proteins in SDS gels. Proteins stained without fixation can be used for zymography (ingel enzyme activity) assays. Stained proteins can also be eluted from gels and used for further analysis. The stain does not alter protein structure and does not interfere with mass spectrometry. In addition, staining does not iterfere with the transfer of proteins to membranes, allowing visualization before preceding with Western blotting. Ex/Em = 300, 490/640 nm

Features & Benefits

- Detects 4-8 ng of protein per mini-gel band
- Staining is complete in less than an hour
- Selectively detects proteins without staining nucleic acids or lipopolysacharide contaminants

Product Code	Description	Size
S 5942	SYPRO Tangerine Gel Stain	1 kit

SYPRO Orange Gel Stain

Molecular Probes' SYPRO Orange protein gel stain provides fast, simple, sensitive staining of proteins in electrophoretic gels.

Features & Benefits

- Detect 4-8 ng of protein per mini-gel band
- Minimize protein-to-protein variability through increased staining consistency and eliminate negative staining
- Fluorescence intensity is linear with protein quantity over three orders of magnitude, much broader than Coomassie or Silver Staining can provide

Product Code	Description	Size
<u>S 5692</u>	SYPRO Orange Gel Stain	1 kit

SYPRO Red Gel Stain

Molecular Probes' SYPRO Red protein gel stain provides fast, simple, sensitive staining of proteins in electrophoretic gels.

Features & Benefits

- Detect 4-8 ng of protein per mini-gel band
- Minimize protein-to-protein variability through increased staining consistency and eliminate negative staining
- Fluorescence intensity is linear with protein quantity over three orders of magnitude, much broader than Coomassie or Silver Staining can provide

Product Code	Description	Size
<u>S 5817</u>	SYPRO Red Gel Stain	1 kit

SYPRO Protein Gel Starter Kit

The SYPRO gel starter kit includes three fluorescent protein stains (SYPRO Orange, SYPRO Red, and SYPRO Tangerine) as well as a SYPRO photographic filter. Proteins as small as 6.5 kDa can be detected with no observed staining of liposaccharides or nucleic acids.

Product Code	Description	Size
<u>SY0100</u>	SYPRO Protein Gel Starter Kit	1 kit

Alcian Blue 8GX

Alcian Blue 8GX is suitable for detection of glycoproteins on nitrocellulose and in PAGE gels. Alcian Blue 8GX reacts with sulfate and carboxylated functional groups. The interaction of Alcian Blue (cationic) and polyanionic glycoproteins is influenced by pH; both sulfate and carboxylate groups will react at pH 2.5, but sulfate groups alone react at pH 1.

Product Code	Description	Size
<u>A 3157</u>	Alcian Blue 8GX	10 g 25 g

Amido Black Staining Solution, 2X concentrate

Amido Black Staining Solution is designed for rapid staining of protein bands on nitrocellulose membranes. Amido Black Staining Solution facilitates visualization of low concentration proteins with a low background. Proteins can be easily destained with 25% (v/v) isopropanol and 10% (v/v) acetic acid for further analysis. Amido black is also compatible with PAGE gels. Each bottle of 2X concentrate dilutes to 500 ml with water.

Product Code	Description	Size
<u>A 8181</u>	Amido Black Staining Solution 2x Concentrate	1 Btl

Eosin Y

Eosin Yellowish reversibly stains peptides and proteins dark red following SDS-PAGE. Proteins can then be recovered from the gel and further characterized.

Product Code	Description	Size
<u>45235</u>	Eosin Y	25 g
		100 g

Fast Green FCF

Fast Green FCF is used for protein detection and quantitation in native PAGE, SDS-PAGE, and IEF gels. Following staining, protein concentration can be measured at 625 nm. Fast Green staining is linear over a wider range of detection than Brilliant Blue R. It is especially well suited to IEF because it does not bind to ampholytes.

Product Code	Description	Size
F 7252	Fast Green FCF	5 g
		25 g

Fluorescamine®

This reagent reacts readily with primary amino groups in amino acids and peptides to form highly-fluorescent compounds.

Product Code	Description	Size
F 9015	Fluorescamine	25 mg 100 mg 1 g

Glycoprotein Detection Kit

The glycoprotein detection kit is designed to selectively stain glycoproteins in polyacrylamide gels and membranes using a modification of the Periodic Acid-Schiff (PAS) methods. Staining of sugar moieties of glycoproteins yields magenta bands with a colorless background. The detection limits using this kit are 25-100 ng of carbohydrate per band. Each kit contains sufficient materials for 10 mini-gels (10 x 10 cm).

Product Code	Description	Size
GLYCO-PRO	Glycoprotein Detection Kit	1 kit

E 3402

Electrophoresis in Practice, 3rd Edition

R. Westermeier, Wiley-VCH, 2001, 368 pp., hard cover



P 4742

Protein Protocols Handbook, 2nd Edition

Walker, J.M., Humana Press, 2002, 809 pp., comb bound





P 2481 2-D Proteome **Analysis Protocols** Link, A.J., Humana Press, 1998, 608 pp., hard cover



Z36,949-7

Strategies for Protein Purification and Characterization: A Laboratory **Course Manual**

D.R. Marshal, et al., Cold Spring Harbor Laboratory Press, 1996, 396 pp., comb bound

Gold, Colloidal

As a widely used protein and polysaccharide marker for electron microscopy, colloidal gold is a useful general stain for proteins immobilized on nitrocellulose membranes following PAGE.

Product Code	Description	Size
<u>50755</u>	Gold, Colloidal	500 ml

Oil Red O

Oil Red O stain is suitable as a lipid/lipoprotein stain on cellulose acetate. A 2% stock solution in methanol is prepared and diluted with sodium hydroxide immediately before staining. The membrane is stained for approximately one hour and then destained in a methanol glycerol solution.

Product Code	Description	Size
<u>O 9755</u>	Oil Red O	25 g 100 g

Ponceau S

Ponceau S (sodium salt) may be used to prepare a stain for rapid reversible detection of protein bands on nitrocellulose or PVDF membranes (Western blotting), as well as on cellulose acetate membranes. Common stain formulations include 0.1% (w/v) Ponceau S in 5% acetic acid or 2% (w/v) Ponceau S in 30% TCA and 30% sulfosalicylic acid.

Product Code	Description	Size
<u>P 3504</u>	Ponceau S	10 g 50 g
		100 g

Ponceau S Solution

Ponceau S Solution is a ready-to-use, reversible staining solution designed for rapid (5 minute) staining of protein bands on nitrocellulose or PVDF membranes (Western blots), as well as cellulose acetate membranes. Ponceau S stain is easily reversed with water washes, facilitating subsequent immunological detection.

Product Code	Description	Size
P 7170	Ponceau S Solution	1 L

Reversible Protein Detection Kit for Membranes and Polyacrylamide Gels

The Reversible Protein Detection Kit for Membranes and Polyacrylamide Gels is a unique protein detection product for visualization of proteins on nylon, nitrocellulose, and PVDF membranes, as well as PAGE gels with sensitivity comparable to that of Coomassie stains. Many other stains produce high backgrounds on nylon membranes due to strong charge interactions. Application of activated R-PROB produces lavenderstained protein bands, which are easily destained (reversed) with an EDTA solution. Each kit is sufficient for 200 applications (small blots on 8 x 10 cm PAGE gels).

Product Code	Description	Size
<u>R-PROB</u>	Reversible Protein Detection Kit	1 kit

Stains-All

Stains-All is uniquely suitable for differential staining of nucleic acids and proteins. RNA bands appear bluish-purple, DNA bands appear blue, and proteins appear red. Typically, PAGE gels are stained in the dark and destained through exposure to light. Staining solutions are commonly made by dissolving the dye in formamide and buffer.

Product Code	Description	Size
<u>E 9379</u>	Stains-All	1 g 5 g

Sudan Black B

Sudan Black B is a lipochrome (fat soluble dye) used to stain phospholipids, neutral fats, and sterols.

Product Code	Description	Size
<u>86015</u>	Sudan Black B	25 g
		100 g

Fixing Solution, 5x concentrate

Fixing solution prevents diffusion or loss of proteins prior to staining with a visualization reagent. 1x fixing solution contains 12% (w/v) trichloracetic acid and 3.5% (w/v) sulfosalicylic acid. Suitable for SDS-PAGE, non-denaturing PAGE, and IEF.

Product Code	Description	Size
<u>F 7264</u>	Fixing Solution, 5x concentrate	500 ml 1 L