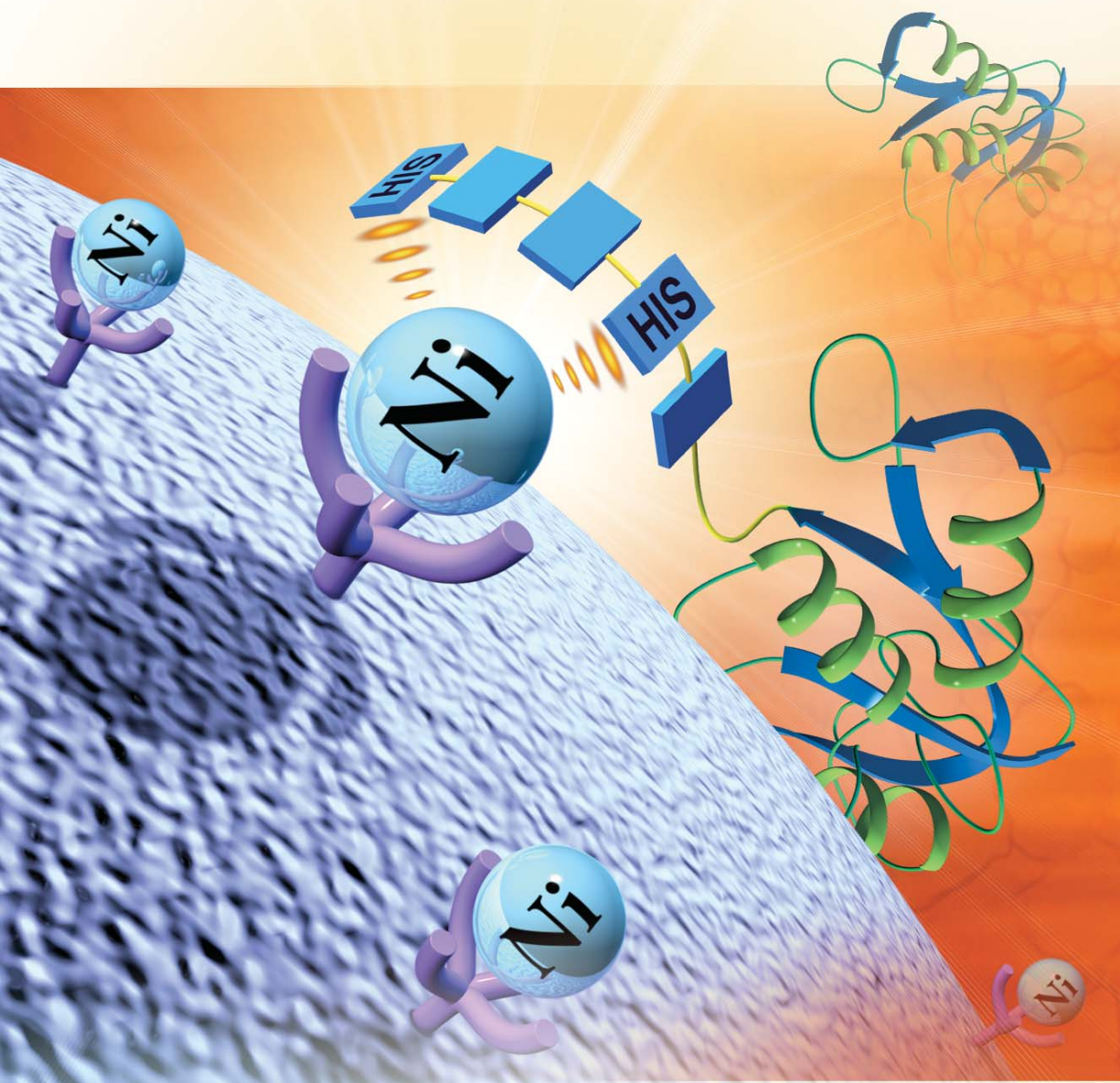


HIS-Select™

Reference & Selection Guide

An Integrated Solution for Purification and Detection



High Selectivity for
Higher Purity

High Binding
Capacity

Reduced Non-
specific Binding

One-step Purification

HIS-Select™ Quick Reference Guide



Product	Nickel Affinity Gel	Nickel High Flow (HF) Affinity Gel	Cobalt Affinity Gel	EZview™ Red Nickel Affinity Gel	Nickel Affinity Gel Cartridges	Nickel High Flow (HF) Affinity Gel Cartridge, 1.25 ml	Nickel High Flow (HF) Affinity Gel Cartridge, 6.4 ml
Product Number	P 6611	H 0537	H 8162	E 3528	H 8286	H 7788	H 7163
Package Sizes	5 ml, 25 ml, 100 ml, 500 ml	10 ml, 25 ml, 100 ml, 500 ml	5 ml, 25 ml, 100 ml	1 ml, 5 x 1 ml	5 ea	1 ea, 5 ea	1 ea, 5 ea
Application	<ul style="list-style-type: none"> • Gravity Flow Column • Small-Med. Scale 	<ul style="list-style-type: none"> • FPLC® • Production Scale 	<ul style="list-style-type: none"> • Gravity Flow Column • Small-Med. Scale 	<ul style="list-style-type: none"> • Mini Prep Pull-Downs 	<ul style="list-style-type: none"> • LPLC • MPLC • Syringe Purification • Small Scale 	<ul style="list-style-type: none"> • LPLC • MPLC • FPLC • Syringe purification 	<ul style="list-style-type: none"> • LPLC • MPLC • FPLC • Syringe purification
Scale per Sample	100 µg – 10 g	100 µg – 100 g	100 µg – 10 g	10 – 500 µg per pull-down	15 – 20 mg	Up to 18 mg per sample	Up to 96 mg per sample
Binding Capacity	15 mg/ml	15 mg/ml	15 mg/ml	15 mg/ml	15 mg/cartridge	15 mg/ml	15 mg/ml
Matrix	6% Beaded Agarose	6% Beaded Agarose, Highly Cross-linked	6% Beaded Agarose	6% Beaded Agarose	6% Beaded Agarose	6% Beaded Agarose, Highly cross-linked	6% Beaded Agarose, Highly cross-linked
Bead Size	45 – 165 µm	45 – 165 µm	45 – 165 µm	45 – 165 µm	45 – 165 µm	45-165 µm	45-165 µm
Exclusion Limit (MW) or Pore Size	4 x 10 ⁶ Da	4 x 10 ⁶ Da	4 x 10 ⁶ Da	4 x 10 ⁶ Da	4 x 10 ⁶ Da	4 x 10 ⁶ Da	4 x 10 ⁶ Da
Max. Linear Flow Rate (Max. pressure)	150 cm/hr (5 psi)	3,000 cm/hr (200 psi)	150 cm/hr (5 psi)	N/A	150 cm/hr (5 psi)	4 ml/min	20 ml/min
Recommended Flow Rate for 1 x 2 cm Column	1 ml/min	5 ml/min	1 ml/min	N/A	1 ml/min	1 ml/min	5 ml/min
Recommended Binding Time/Speed	N/A	N/A	N/A	1 hr @ 4 °C	N/A	N/A	N/A
Optimal pH Stability	3-10	3-10	3-10	3-10	3-10	3-10	3-10
Physical Form	50% suspension in 30% ethanol	50% suspension in 30% ethanol	50% suspension in 30% ethanol	50% suspension in 30% ethanol	1.25 ml pre-packed cartridge in 30% ethanol	Pre-packed cartridge in 30% ethanol	Pre-packed cartridge in 30% ethanol
Antimicrobial Agent	30% ethanol	30% ethanol	30% ethanol	30% ethanol	30% ethanol	30% ethanol	30% ethanol
Storage	2-8 °C	2-8 °C	2-8 °C	2-8 °C	2-8 °C	2-8 °C	2-8 °C
Recommended Imidazole Conc. for Load/Wash	0-10 mM	0-10 mM	0-10 mM	0-10 mM	0-10 mM	0-10 mM	0-10 mM
Recommended Imidazole Conc. for Elution	250 mM (H 5413)	250 mM (H 5413)	250 mM (H 5413)	250 mM (H 5413)	250 mM (H 5413)	250 mM (H 5413)	250 mM (H 5413)

For more information, please visit



see our automation protocols at sigma-aldrich.com/automation



COMING SOON!

Nickel Spin Columns	Nickel Magnetic Beads	High Sensitivity (HS) Nickel Coated Plates	High Capacity (HC) Nickel Coated Plates 96-well	Filter Plates 96-well	iLAP™ High Capacity (HC) Nickel Coated Plates 96-well	HIS-Select™ iLAP columns	HIS-Select Ni-ABC-HRP
H 7787	H 1786	S 5688 & S 9191	S 5563	H 0413	H 9412	H 9913	N 1412
10 ea, 50 ea	2 x 1 ml, 5 x 1 ml	1 ea, 5 ea	1 ea, 5 x 1 ea	1 ea, 5 ea	1 ea, 5 x 1 ea	1 ea, 25 ea	250 µg protein (HRP)
<ul style="list-style-type: none"> • Mini Prep • Process by Centrifugation or Vacuum 	<ul style="list-style-type: none"> • Rapid Mini Prep Magnetic Pull-Downs • High Throughput Screening • Compatible with Robotics when Used in 96-Well Plates 	<ul style="list-style-type: none"> • High Throughput Screening (ELISA) • Compatible with Robotics 	<ul style="list-style-type: none"> • High Throughput, High Capacity Screening • Multi-Sample Mini Prep • In-Well Protein Assays • Compatible with Robotics 	<ul style="list-style-type: none"> • Small Scale • High Throughput Screening • Process by Centrifugation or Vacuum • Compatible with Robotics 	<ul style="list-style-type: none"> • Direct Cell Lysis and Purification of Target Protein in Well • High Throughput, High Capacity Screening • Multi-Sample Mini Prep • In-Well Protein Assays • Compatible with Robotics 	<ul style="list-style-type: none"> • Direct Cell Lysis and Purification of Target Protein in Gravity Flow Column 	<ul style="list-style-type: none"> • Western Blot • Dot Blot • ELISA
<ul style="list-style-type: none"> • 150 µg • 600 µl/load 	10 µg – 15 mg per pull-down	<ul style="list-style-type: none"> • 1 ng – 1 µg/well • 96-well: 200 µl/well 	<ul style="list-style-type: none"> • 1 – 4 µg/well • 200 µl/well 	<ul style="list-style-type: none"> • 1 mg/well • 1 ml/load 	<ul style="list-style-type: none"> • 1 – 4 µg/well • 96-well: 200 µl/well 	Up to 1 mg	Up to 100 blots or 10,000 wells
150 µg/column	15 mg/ml	Sensitivity: ≤1 ng/well	≥4 µg/well	1 mg/well	≥4 µg/well	1 mg per 5 ml culture	Detection limit of 5 ng by dot blot
Silica Particles (Porous, Spherical)	Paramagnetic Iron Impregnated 6% Beaded Agarose	Clear Polystyrene Plate; Proprietary Coating	Clear Polystyrene Plate; Proprietary Coating	Silica Particles (Porous, Spherical)	Clear Polystyrene Plate; Proprietary Coatings for lysis and purification	Proprietary resin and buffers for lysis and purification	Nickel Avidin-Biotin-Complex Peroxidase Conjugate
Approx. 20 µm	20 – 75 µm	N/A	N/A	Approx. 20 µm	N/A	45-165 µm	N/A
0.1 µm (approx. 10 x 10 ⁶ Da)	4 x 10 ⁶ Da	N/A	At least 300 kDa	0.1 µm (approx. 10 x 10 ⁶ Da)	At least 300 kDa	4 x 10 ⁶ Da	N/A
N/A	N/A	N/A	N/A	N/A	N/A	150 cm/hr (5 psi)	N/A
N/A	N/A	N/A	N/A	N/A	N/A	1 ml/min	N/A
Equivalent to approx. 80 x g Centrifugation	0.5 hr @ 25 °C	1 hr @ 25 °C	4 hr @ 25 °C	Equivalent to approx. 270 x g Centrifugation	1-4 hr @ 25 °C	N/A	Blot: 1.75 hr, Plate: 1.5 hr
3-10	3-10	3-10	3-10	3-10	3-10	3-10	N/A
Pre-packed dry matrix in spin column	50% suspension in 30% ethanol	Clear 96-well-flat bottom, polystyrene strip plate (S 5688) or solid plate format (S 9191)	Clear 96-well polystyrene plates	Pre-packed dry matrix in 96-well filter plate	Clear 96-well flat bottom, polystyrene plates	Pre-packed cartridge containing lyophilized powder/dry resin	Lyophilized powder
N/A	30% ethanol	N/A	chlorhexidine	N/A	N/A	N/A	N/A
2-8 °C	2-8 °C	2-8 °C	2-8 °C	2-8 °C	2-8 °C	-20 °C	-20 °C as supplied, 2-8 °C after reconstitution
0-5 mM	0-10 mM	0-5 mM	0-1 mM	0-5 mM	0-1 mM	5 mM	40 mM
250 mM (H 5413)	250 mM (H 5413)	N/A	200 mM	250 mM (H 5413)	200 mM	250 mM (H 5413)	500 mM

NEW HIS-Select™ High Flow Cartridge (H 7788 & H 7163)

The HIS-Select Cartridge is a pre-packed ready to use cartridge (column) that contains HIS-Select High Flow Nickel Affinity Gel (Product Code H 0537), offering superior selectivity using our HIS-Select Immobilized Metal Affinity Chromatography (IMAC) technology. A highly cross-linked agarose allows higher flow rates and mechanical stability under pressure. This cartridge's luer lock fittings and accompanying adaptors make it suitable for Low Pressure Liquid Chromatography Systems (LPLC), Medium Pressure Liquid Chromatography Systems (MPLC), Fast Pressure Liquid Chromatography (FPLC™), and for use with a syringe. Each cartridge contains 1.25 ml (H 7788) or 6.4 ml (H 7163) of HIS-Select High Flow Nickel Affinity Gel.

NEW HIS-Select iLAP™ 5 ml column (H 9913)

HIS-Select iLAP (Integrated Lysis and Affinity Purification) columns are single-use, disposable columns designed for one-step purification of histidine-tagged proteins directly from a 5 ml bacterial culture. The single-step method uses our innovative iLAP technology, which allows quick and simple colony screening for histidine-tagged recombinant clones. Each column is capable of purifying at least 1 milligram of histidine-tagged protein in less than an hour, resulting in a nearly pure fusion protein preparation following column elution.

NEW HIS-Select Ni-ABC-HRP (N 1412)

HIS-Select Ni-ABC-HRP is used for the detection of histidine-tagged proteins in a variety of standard methods without the use of antibodies. This product is composed of chelated nickel linked to peroxidase via an avidin-biotin complex. This product is designed for use in Western blotting where 50 ng of histidine-tagged protein can be detected as the major band in 10 µg of total protein from an *E. coli* lysate. The product can detect <5 ng of histidine-tagged protein in a dot blot or ELISA assay.

HIS-Select Nickel Affinity Gel (P 6611)

This product is the tool of choice for small to medium scale purification of histidine-tagged recombinant proteins. The unique, non-charged, hydrophilic linkage of the nickel chelate group to the agarose ensures high selectivity for histidine-tagged proteins and reduces non-specific binding of other proteins and is compatible with both denaturing or non-denaturing conditions.

HIS-Select HF Nickel Affinity Gel (H 0537)

HIS-Select HF (High Flow) Nickel Affinity Gel brings the superior selectivity of HIS-Select Immobilized Metal Affinity Chromatography (IMAC) technology to a highly cross-linked agarose designed for higher flow rates and mechanical stability under pressure. This product is designed for production scale purification and FPLC applications. As with other HIS-Select products, the non-charged, hydrophilic linkage of the NTA chelate group to the agarose ensures true one-step purification.

HIS-Select Cobalt Affinity Gel (H 8162)

For those who prefer to use cobalt as the affinity metal ion for histidine-tagged protein purification, this product allows for high purity, low non-specific binding, and high binding capacity for your protein. This product also works well for purification of histidine-tagged proteins in native, denaturing, or mild reducing conditions.

HIS-Select Nickel Cartridge (H 8286)

These pre-packed columns are filled with 1.25 ml of HIS-Select Nickel Affinity Gel (P 6611). The cartridges allow for rapid, small-scale purification of histidine-tagged proteins. Columns contain luer lock fittings that allow syringe purification, or connection to low or medium pressure liquid chromatography systems (5 psi maximum pressure). Plus, each pre-packed column binds ≥15 mg of histidine-tagged recombinant protein.

HIS-Select Spin Columns (H 7787)

Pre-packed and ready-to-use HIS-Select Spin Columns allow for fast and consistent small-scale purification of histidine-tagged protein from crude cell extracts. The Spin Columns contain a matrix of silica particles and use the HIS-Select technology to attain high protein purity and very low non-specific binding. Spin Columns allow for fast, one-step purification of at least 150 µg of histidine-tagged protein in less than 15 minutes.

EZview™ Red HIS-Select HC Nickel Affinity Gel (E 3528)

When performing small scale affinity capture, such as immunoprecipitation, the affinity matrix is difficult to see in the microcentrifuge tubes. Accidental aspiration of the resin leads to quantitative variability in results. The EZview Red Affinity Gel greatly reduces the risk of pellet loss. EZview resins perform as well as conventional non-colored affinity gels, but allow the user to easily differentiate pellet from supernatant. This correlates to more accurate data because less target protein is lost.

HIS-Select HC Nickel Magnetic Beads (H 1786)

The HIS-Select Magnetic Agarose Beads consist of paramagnetic, immobilized metal-ion affinity chromatography (IMAC) resin, designed for use in automated and small-scale affinity capture (molecular pull-down) purification of histidine-tagged proteins. The magnetic properties make manipulations such as repetitive washings and recovery of bound protein from the beads quicker and easier. This leads to greater speed, high selectivity, experimental reproducibility, and more accurate quantification of your protein of interest.

HIS-Select High Sensitivity (HS) Nickel Coated Plates (S 5688 & S 9191)

HIS-Select High Sensitivity (HS) Nickel Coated Plates are designed for highly accurate low-level detection of histidine-tagged proteins. The captured proteins can be detected using standard enzyme-linked assay (ELISA) techniques and are suitable for high throughput applications. These plates are manufactured by utilizing a unique non-charged hydrophilic linkage for the nickel-chelate, are pretreated to reduce non-specific binding, and are sensitive enough to capture as little as 1 ng/well of histidine-tagged protein. These plates are supplied in a 96-well strip plate (S 5688) or single plate format (S 9191).

HIS-Select High Capacity (HC) Nickel Coated Plates (S 5563)

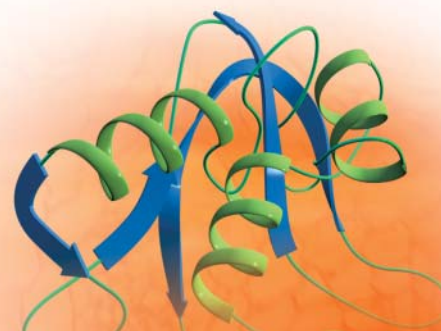
HIS-Select High Capacity (HC) Nickel Coated Plates are coated with a proprietary, high-density nickel chelate matrix. This coating allows for greater per-well binding capacity (≥4 µg/well) than any other commercial histidine-binding plate and still demonstrates low non-specific binding. These plates allow for the in-well quantification of bound protein and are suitable for applications including: Expression Screening, PAGE, MALDI-MS, and High Throughput Purification.

HIS-Select iLAP HC Nickel Coated Plate (H 9412)

Integrated Lysis and Affinity Purification (iLAP) plates are innovative products, which combines cell lysis and histidine-tagged protein purification steps into one. These plates are coated with a proprietary, high-density nickel chelate matrix along with an additional coating for simultaneous and highly efficient cell lysis and target protein extraction, eliminating the need to harvest bacterial cultures before processing. The nickel chelate coating is highly selective for histidine-tagged proteins, reducing non-specific binding. The High Capacity feature of the 96-well plates enables capture of ≥4 µg of histidine-tagged protein per well, making these plates ideal for rapid colony screening, protein:protein interaction assays, and automation systems. The resulting purified protein is compatible with standard protein assays, SDS-PAGE, Western blotting, and MALDI.

HIS-Select Filter Plate (H 0413)

The filter plate is an immobilized metal-ion affinity chromatography (IMAC) product that incorporates the speed, consistency, and convenience of spin columns into a multi-sample format. This pre-packed 96-sample plate has wells that contain 20 µm, spherical silica particles (100 nm pore size), which are derivitized with a nickel chelate linkage. The particles allow for small scale histidine-tagged protein purification from crude cell extracts in less than 15 minutes. The filter plates are also compatible with centrifugation, vacuum-manifolds, and robotics systems.



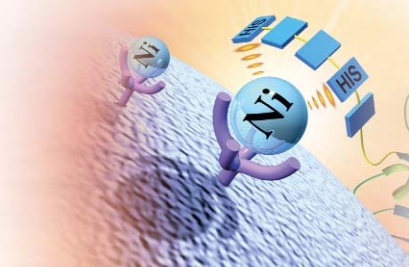
HIS-Select™ Purification Kits

Product	HIS-Select™ M Affinity Capture Kit	HIS-Select M Purification Kit	HIS-Select Y Purification Kit
Product Number	EHM-1	HMP-1	HYP-1
Package Sizes	1 Kit, sufficient for 50 samples	1 Kit, 5 individual columns	1 Kit, 5 individual columns
Application	Provides all the components needed for the extraction and rapid small-scale affinity capture of histidine-tagged proteins from mammalian cells	Complete kit for lysis, protein extraction, and affinity purification of histidine-tagged proteins from mammalian cells	Complete kit for lysis, extraction, and purification of histidine-tagged proteins from yeast
Scale per Sample	40 µl of gel suspension (20 µl of packed gel)	1 ml per column	1 ml per column
Binding Capacity	≥15 mg/ml	≥15 mg/ml	≥15 mg/ml
Affinity Gel Type *see respective resin product number for additional information	E 3528	P 6611	P 6611
Components	<ul style="list-style-type: none"> • CelLytic™ M Lysis Buffer (C 2978) • 5X Phosphate Buffer: 250 mM Sodium Phosphate, pH 8, with 1.5.M NaCl • 2.5% CTAB • 2.5 M Imidazole, pH 8.0 • 5% TWEEN® 20 • EZview™ Red HIS-Select HC Nickel Affinity Gel (E 3528) • Protease Inhibitor Cocktail (P 8340) 	<ul style="list-style-type: none"> • CelLytic M Lysis Buffer (C 2978) • 5X Phosphate Buffer: 250 mM Sodium Phosphate, pH 8, with 1.5.M NaCl; • 2.5% CTAB • 2.5 M Imidazole, pH 8.0 • 5% TWEEN 20 • HIS-Select Nickel HC Affinity Gel (P 6611) • 5 Polypropylene Chromatography Columns 	<ul style="list-style-type: none"> • CelLytic Y Lysis Buffer (C 4482) • 5X Phosphate Buffer: 250 mM Sodium Phosphate, pH 8, with 1.5.M NaCl • 2.5% CTAB • 2.5 M Imidazole, pH 8.0 • 5% TWEEN 20 • HIS-Select Nickel HC Affinity Gel (P 6611) • 5 Polypropylene Chromatography Columns
Description	This kit provides all the components needed for the extraction and rapid, small-scale affinity capture of histidine-tagged proteins from mammalian cells. The CelLytic M lysis buffer allows for quick and effective lysis of both adherent and suspension cells while the EZview Red HIS-Select Nickel Affinity Gel (E 3528) provides rapid affinity purification of histidine-tagged proteins with high selectivity and very low non-specific binding of other proteins. Plus, the red color allows for less pellet loss during aspiration when performing immunoprecipitation or molecular pull-down experiments. The kit is sufficient for 50 affinity capture purifications.	This kit provides all the components needed for the extraction and rapid affinity purification of histidine-tagged proteins from mammalian cells. The CelLytic M lysis buffer allows for quick and effective lysis of both adherent and suspension cells while the HIS-Select Nickel Affinity Gel (P 6611) provides rapid affinity purification of histidine-tagged proteins with high selectivity and very low non-specific binding of other proteins. This kit allows for one-step purification of ≥15 mg of histidine-tagged protein per ml of affinity gel. The kit is sufficient for 5 x 1 ml affinity purifications.	This kit provides all the components needed for the extraction and rapid affinity purification of histidine-tagged proteins from yeast cells. The CelLytic Y lysis buffer allows for quick and effective lysis of yeast cells while the HIS-Select Nickel Affinity Gel (P 6611) provides rapid affinity purification of histidine-tagged proteins with high selectivity and very low non-specific binding of other proteins. This kit allows for one-step purification of ≥15 mg of histidine-tagged protein per ml of affinity gel. The kit is sufficient for 5 x 1 ml affinity purifications.

HIS-Select™ Buffers

Product	HIS-Select Wash Buffer	HIS-Select Elution Buffer	HIS-Select Wash & Elution Buffer Kit
Product Number	H 5288	H 5413	HS0100
Package Sizes	500 ml, 1 L	250 ml, 500 ml	500 ml wash buffer, 250 ml elution buffer
Application	Optimized to reduce non-specific binding of proteins during wash steps when purifying histidine-containing proteins	Elute histidine-containing target proteins from HIS-Select purification products or other immobilized metal affinity chromatography (IMAC) products.	Optimized to reduce binding of non-specific proteins during purification and to allow the specific elution of histidine-containing target protein(s).
Components	<ul style="list-style-type: none"> • 10 mM imidazole • 0.3 M sodium chloride • 50 mM sodium phosphate, pH 8.0 	<ul style="list-style-type: none"> • 250 mM imidazole • 0.3 M sodium chloride • 50 mM sodium phosphate, pH 8.0 	<ul style="list-style-type: none"> • Contains a 10 mM Imidazole Wash Buffer and 250 mM Imidazole Elution Buffer. • Both buffers also contain 0.3 M sodium chloride, 50 mM sodium phosphate, pH 8.0
Description	HIS-Select Wash Buffer is a pre-made solution of 10 mM imidazole (in 0.3 M sodium chloride, and 50 mM sodium phosphate, pH 8.0) that is optimized to reduce non-specific binding of proteins during wash steps when purifying histidine-containing proteins using HIS-Select purification products or other immobilized metal affinity chromatography (IMAC) products.	HIS-Select Elution Buffer is a pre-made solution of 250 mM imidazole (in 0.3 M sodium chloride, and 50 mM sodium phosphate, pH 8.0) that is optimized to elute histidine-containing target proteins from HIS-Select purification products or other immobilized metal affinity chromatography (IMAC) products.	The HIS-Select Buffer Kit contains a 10 mM imidazole Wash Buffer (H 5288) and a 250 mM imidazole Elution Buffer (H 5413) for use in procedures for purification of recombinant proteins with histidine or other metal affinity tags. The buffers are designed for use with all HIS-Select Products and other immobilized metal affinity chromatography (IMAC) products. The wash buffer is optimized to reduce binding of non-specific proteins during purification. The elution buffer is optimized for elution of the bound histidine-tagged protein(s). The buffers are sold individually or in this kit. Both the Wash Buffer and Elution Buffer contain 0.3 M sodium chloride, 50 mM sodium phosphate, pH 8.0.

For more information, please visit
our website at sigma-aldrich.com/HISselect



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