Glycoproteomics Selection Guide — Deglycosylation

	Deglycosylation					
Product	GlycoProfile I In-Gel Deglycosylation Kit	GlycoProfile II Ezymatic In-Solution Deglycosylation Kit	GlycoProfile IV Chemical Deglycosylation Kit	Native Enzymatic Deglycosylation Kit	Enzymatic Deglycosylation Kit	**Product**
Product Number	**PP0200**	**PP0201**	**PP0510**	**NDEGLY**	**EDEGLY**	**Product Number**
Recommended Sample Size	PAGE gel containing up to 5 mg of glycoprotein	Solutions containing 1-2 mg of glycoprotein	Solutions containing 1-2 mg of glycoprotein	Solutions containing up to 200 mg of glycoprotein	Solutions containing up to 200 mg of glycoprotein	**Recommended Sample Size**
Sample Scale	Minimum of 10 samples	Minimum of 20 reactions	Minimum of 10 reactions	Minimum of 10 reactions	Minimum of 10 reactions	**Sample Scale**
Product Description and Applications	This kit includes PNGase F, the most common enzyme used for removal of N-linked glycans, and trypsin for tryptic digestion of the core protein. The conditions are optimized to provide a convenient and reproducible method of *in-gel* removal of N-linked glycans from glycoproteins and *digestion* of the core protein with trypsin. The samples can then be desalted and concentrated for analysis by MALDI-TOF-MS or ESI-MS. This kit also works for N-deglycosylation *in solution.*	This kit includes PNGase F, which removes N-linked glycans and leaves the protein core essentially intact, except for the conversion of Asn to Asp. The released N-linked glycans are amenable for compositional, structural, and other analyses, while the core protein can be analyzed by MS. The conditions are optimized to provide a convenient and reproducible method to remove N-linked glycans from glycoproteins *in solution* for subsequent MALDI-TOF MS analysis without interference from any of the reaction components.	This kit utilizes anhydrous trifluoromethanesulfonic acid (TFMS) to remove O- and N-linked glycans from glycoproteins (except the innermost N-linked GlcNAc or GalNAc). There is minimal degradation of the protein core, which can then be recovered for analysis. The conditions have been optimized to deglycosylate simple or low molecular weight glycoproteins in 30 min without scavenger. For complex or high molecular weight, non-mammalian glycoproteins, anhydrous anisole is provided as scavenger to ensure the highest protein yield as possible.	The NDEGLY kit consists of three types of endoglycosidases (F1, F2, and F3) and is designed for the removal of N-linked oligosaccharides from PNGase F-resistant native proteins. Endoglycosidases F1, F2, and F3 are less sensitive to protein conformation than PNGase F and are more suitable for removal of all classes of N-linked oligosaccharides without prior denaturation of the protein.	The EDEGLY kit contains all enzymes and reagents needed to completely remove all N- and simple O-linked carbohydrates from glycoproteins under native or denaturing conditions. Additional enzymes and reagents are included for cleavage of complex core 2 O-linked carbohydrates including those containing polylactosamine. The core protein remains intact and can be amenable for structure and function studies or MS analysis.	**Product Description and Applications**
Features and Benefits	• In-gel deglycosylation and digestion avoids extra preparative steps • Highly purified enzymes to prevent unwanted activities and products	• Reagents are optimized for direct MS analysis without extra clean-up step • Highly purified enzymes to prevent unwanted activities and products	• Complete deglycosylation in as short as 30 min for increased throughput • Minimal degradation of protein core for more reliable MS data	• Glycoproteins are deglycosylated in their native state • Core protein is suitable for structure function studies or MS analysis	• Single reaction at neutral pH eliminates pH adjustment for each glycosidase reaction • Native or denaturing conditions allow flexibility for downstream process	**Features and Benefits**
Components	• Destaining Solution, reconstitutes to 10 ml **(D0316)** • Proteomics Grade PNGase F, 50 units **(P7367)** • Proteomics Grade Trypsin, 20 µg **(P6567)** • Trypsin Solubilization Reagent, 1 ml **(T2073)** • Trypsin Reaction Buffer, reconstitutes to 11 ml **(R3527)** • Invertase Glycoprotein standard, 0.5 mg **(I0408)** • Peptide Extraction Solution, 10 ml **(P0743)** • Acetonitrile, Biotech Grade, 50 ml **(49,444-5)**	• Proteomics Grade PNGase F, 50 units **(P7367)** • Ribonuclease B glycoprotein standard, 0.5 mg **(P7884)** • 10X Reaction Buffer, 158 mg of ammonium bicarbonate **(I1283)** • Octyl β-D-glucopyranoside, 100 mg **(O9882)** • 2-Mercaptoethanol, 0.90 ml **(M3148)**	• Trifluoromethanesulfonic acid, anhydrous, 5 x 1.0 g **(34,781-7)** • Ribonuclease B Glycoprotein Standard, 3 x 1.0 mg **(R1153)** • Pyridine Solution, 60%, 1 x 10 ml **(P5496)** • Bromophenol Blue Solution, 0.2%, 1 x 0.5 ml **(B1560)** • Anisole, anhydrous, 5 x 1 ml **(29,629-5)** • Reaction Vials with Caps, 10 each **(27265/27273)**	• Endoglycosidase F1, 0.3 unit **(E9762)** • Endoglycosidase F2, 0.1 unit **(E0639)** • Endoglycosidase F3, 0.1 unit **(E2264)** • Endoglycosidase F1 Reaction Buffer, 200 µl **(R9025)** • Endoglycosidase F2 and F3 Reaction Buffer, 200 µl **(R9150)**	• PNGase F, 20 µl, 0.3 unit (P2619) • O-Glycosidase, 20 µl (G1163) • α-2(3,6,8,9)-Neuraminidase, 20 µl (N8271) • Fetuin Control, 10 mg/ml solution, 0.5 mg (F4301) • 5X Reaction Buffer, 0.2 ml (R2651) • Denaturation Solution, 0.1 ml (D6439) • Triton X-100, 15% Solution, 0.1 ml (T3319) • β-1,4-Galactosidase, 20 µl (G0413) • β-N-Acetylglucosaminidase, 20 µl (A6805)	**Components**
Storage Temperature	2-8 °C	2-8 °C	2-8 °C	4 °C	4 °C	**Storage Temperature**
Shelf Life of Unused Product	Minimum of 1 year	Minimum of 1 year	Minimum of 1 year	Minimum of 1 year	Minimum of 1 year	**Shelf Life of Unused Product**

Glycoproteomics Selection Guide — Labeling and Detection, Quantitation

	Labeling and Detection				Quantitation	
Product	GlycoProfile 2-AA Labeling Kit	GlycoProfile 2-AB Labeling Kit	GlycoProfile III Fluorescent Glycoprotein Detection Kit	Glycoprotein Detection Kit	Sialic Acid Quantification Kit	**Product**
Product Number	**PP0530**	**PP0520**	**PP0300**	**GLYCOPRO**	**SIALICQ**	**Product Number**
Recommended Sample Size	Solutions containing 100 pmol – 50 nmol of purified glycans	Solutions containing 100 pmol – 50 nmol of purified glycans	Proteins bands containing 10-500 ng of carbohydrates	Protein bands containing 25-100 ng of carbohydrates	Solutions containing 1-200 nmoles of NANA	**Recommended Sample Size**
Sample Scale	Each kit is sufficient for labeling up to 36 samples	Each kit is sufficient for labeling up to 36 samples	For 10 minigels or membranes of the same sizes	For 10 minigels or 5 large gels or membranes of the same sizes	Minimum 25 reactions, including control samples	**Sample Scale**
Product Description and Applications	This kit is optimized for efficient labeling of N-linked, O-linked, and glycosylphosphatidylinositol (GPI) anchored glycans (anthranilic acid; 2-AA). This small fluorophore increases the spectral absorption of glycans, improving the detection of labeled glycans by HPAE, HPLC, and ESI-MS. Binding is robust and dye-glycan conjugates are stable, with no degradation analysis. Although 2-AA and 2-AB are suitable for the same applications, 2-AA is more sensitive than 2-AB and more suitable for SDS-PAGE.	The GlycoProfile 2-AB kit is optimized for efficient labeling of N-linked, O-linked, and glycosyl-phosphatidylinositol (GPI) anchored glycans with 2-aminobenzamide (2-AB). This small fluorophore increases the spectral absorption of glycans, improving the detection of labeled glycans by HPAE, HPLC, and ESI-MS. Binding is robust and dye-glycan conjugates are stable, with no degradation analysis. Although 2-AA and 2-AB are suitable for the same applications, 2-AB is less sensitive than 2-AA.	The Glycoprofile III is a highly selective and sensitive method for the fluorescent detection of glycosylated proteins utilizing standard UV-transillumination directly on PAGE gels without blotting or use of antibodies. Periodic acid is used to oxidize the carbohydrates on the proteins into aldehydes, which then form a stable conjugate with a hydrazide derivative of a fluorescent dye. This allows for specific and sensitive detection of the glycoproteins in gels or in PVDF membranes.	The Glycoprotein Detection Kit is designed as a convenient, reliable method of detecting sugar moieties of glycoproteins in PAGE gels or Western blotting membranes using a modification of the Periodic Acid-Schiff (PAS) reaction. The periodic acid-Schiff reagent stains vicinal diol groups found mainly in peripheral sugars and sialic acids, and is used as a general glycoprotein stain that produces magenta bands with a colorless background.	The Sialic Acid Quantification kit provides a rapid and accurate determination of total N-acetylneuraminic acid (NANA; sialic acid). NANA is released from glycoconjugates using neuraminidase. The kit uses α(2→3,6,8,9) neuraminidase to cleave all NANA linkages, including α(2→8) and α(2→9) linkages, as well as branched NANA, for the most accurate determination of total sialic acid content.	**Product Description and Applications**
Features and Benefits	• Suitable for downstream glycan analysis by HPAE, HPLC, and ESI-MS • More sensitive than 2-AB and more suitable for labeling glycoproteins in SDS-PAGE	• Suitable for downstream glycan analysis by HPAE, HPLC, and ESI-MS	• Superior selectivity enables more accurate detection of glycoproteins • Excellent sensitivity allows detection of 10-500 ng of carbohydrate	• Convenient and reliable detection of glycoproteins • High sensitivity detects as little as 25-100 ng of carbohydrate	• Rapid and accurate method for total sialic acid content • High sensitivity allows measurement down to 1-200 nmoles of sialic acid	**Features and Benefits**
Components	• 2-AA (Anthranilic Acid), 2 x 6 mg **(A6729)** • DMSO (Dimethyl sulfoxide), 2 x 350 ml **(D4942)** • Acetic acid, Glacial, 2 x 200 ml **(A9353)** • Reductant (Sodium Cyanoborohydride), 2 x 6 mg **(R5153)**	• 2-AB (2-Aminobenzamide), 2 x 5 mg **(A9478)** • DMSO (Dimethyl sulfoxide), 2 x 350 ml **(D4942)** • Acetic acid, Glacial, 2 x 200 ml **(A9353)** • Reductant (Sodium Cyanoborohydride), 2 x 6 mg **(R5153)**	• ProteoProfile PTM Marker, 100 ml **(P1745, also available separately)** • Oxidation Reagent, 10X periodic acid solution in water, 250 ml **(O2014)** • Glycoprotein Staining Reagent, 100X dansyl hydrazide dye concentrate in acetonitrile, 10 ml **(G8418)** • Staining Buffer, 2 x 500 ml **(W2014)**	• Oxidation Reagent (Periodic Acid), reconstitutes to 1000 ml solution **(O0258)** • Reduction Reagent (Sodium Metabisulfite), reconstitutes to 1000 ml **(R0764)** • Schiff's Reagent, Fuchsin-Sulfite Reagent, ___ ml **(S5133)** • Peroxidase from Horseradish **(P2075)**	• N-Acetylneuraminic Acid Aldolase, 25 ml **(A0849)** • α-(2→3,6,8,9)-Neuraminidase (Sialase A), 25 ml **(N8271)** • L-Lactic Dehydrogenase, 25 ml **(L9889)** • β-NADH, Disodium Salt, 3 x 5 mg **(N8129)** • Fetuin, Bovine, 0.5 mg **(F4301)** • N-Acetylneuraminic Acid (NANA), 0.01M, 200 ml **(A0974)** • Tris-HCl, 1.0M, pH 7.5, 1 ml **(T3946)** • Sialidase Buffer (250 mM Sodium phosphate, pH 5.0, 200 ml **(S7189)**	**Components**
Storage Temperature	Room temperature	Room temperature	2-8 °C	2-8 °C	2-8 °C	**Storage Temperature**
Shelf Life of Unused Product	Minimum of 1 year	Minimum of 1 year. 2-AB is light sensitive and stored in the dark.	Minimum of 1 year	Minimum of 1 year	Minimum of 1 year	**Shelf Life of Unused Product**