

A Spin-Tip Enrichment Strategy for Simultaneous Analysis of N-Glycopeptides and Phosphopeptides from Human Pancreatic Tissues

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Materials

Name	Company	Catalog Number	Comments
Acetic Acid, Glacial (Certified ACS)	Fisher Scientific	A38S-500	
Acetone (Certified ACS)	Fisher Scientific	A18-1	
Acetonitrile, Optima LC/MS Grade	Fisher Scientific	A955-4	
Ammonium Acetate (Crystalline/Certified ACS)	Fisher Scientific	A637-500	
Ammonium Hydroxide (Certified ACS Plus)	Fisher Scientific	A669-212	
Byonic software	Protein Metrics	n/a	Commercial software used for glycoproteomic analysis (https://proteinmetrics.com/byos/)
C18 BEH material	Waters	186002353	Material removed from column and used to pack nano capillaries (pulled to integrate tip used directly in line with instrument inlet)
CAE-Ti-IMAC, 100%	J&K Scientific	2749380-1G	Material used for dual-functional Ti(IV)-IMAC; can also be used for conventional IMAC/conventional phosphopeptide enrichment
Cellcrusher kit	Cellcrusher	n/a	Used for grinding tissue samples into powder before extraction
Eppendorf 5424R Microcentrifuge	Fisher Scientific	05-401-205	For temperature-controlled centrifugation
cOmplete protease inhibitor cocktail tablets	Sigma	11697498001	
DTT, Molecular Grade (DL-Dithiothreitol)	Promega	V3151	Protein reducing agent
Ethanol, 200 proof (100%), USP	Fisher	22-032-601	
Fisherbrand Analog Vortex Mixer	Fisher Scientific	02-215-414	
Fisherbrand Low-Retention Microcentrifuge Tubes (1.5 mL)	Fisher Scientific	02-681-320	

Fisherbrand Low-Retention Microcentrifuge Tubes (2 mL)	Fisher Scientific	02-681-321	
Fisherbrand Model 120 Sonic Dismembrator	Fisher Scientific	FB120110	For sample lysis using ultrasonication
Formic Acid, 99.0+%, Optima LC/MS Grade	Fisher Scientific	A117-50	
Fused silica capillary (75 µm inner diameter, 360 µm outer diameter)	Polymicro Technologies LLC	100 m TSP075375	For in-house pulled and packed columns with integrated emitter
Hydrofluoric acid (48 wt. % in H ₂ O)	Sigma-Aldrich	339261-100ML	Used for opening emitter of pulled capillary column
Iodoacetamide, BioUltra	Sigma	I1149-5G	Protein reducing reagent
MaxQuant software	n/a	n/a	Free software used for phosphoproteomic analysis (https://www.maxquant.org/)
Multi-therm Shaker with heating and cooling	Benchmark Scientific	H5000-HC	Heating block
Oasis HLB 1 cc Vac Cartridge, 10 mg Sorbent per Cartridge, 30 µm, 100/pk	Waters	186000383	Larger-scale cartridge desalting for tryptic digests (loading capacity approximately up to 1 mg each)
OMIX C18 pipette tips, 100 µL tip, 10 - 100 µL elution volume, 1 x 96 tips	Agilent	A57003100	Smaller-scale packed pipette tip for desalting for enrichment elutions
P-2000 Micropipette Puller	Sutter Instrument Co.	P-2000/F	For pulling nano-capillary columns for LC-MS
PhosSTOP phosphatase inhibitor tablets	Sigma	4906845001	
Pierce BCA Protein Assay Kit	Thermo Fisher Scientific	23225	
Pierce Quantitative Colorimetric Peptide Assay	Thermo Fisher Scientific	23275	
PolySAX LP (12 µm, pore size 300 Å)	PolyLC	BMSX1203	Material for strong anion-exchange chromatography used for ERLIC/ conventional glycopeptide enrichment
Potassium Phosphate Monobasic (Crystalline/Certified ACS)	Fisher Scientific	P285-500	
Pressure injection cell with integrated magnetic stirplate	Next Advance	PC77-MAG	For packing nano-capillary columns with stationary phase up to 2500 psi limit
Proteome Discoverer software	Thermo Fisher Scientific	n/a	Commercial software for proteomics analysis (with integrated database searching software nodes) and data visualization (https://www.thermofisher.com/us/en/home/industrial/mass-spectrometry/liquid-chromatography-mass-spectrometry-lc-ms/lc-ms-software/multi-omics-data-analysis/proteome-discoverer-software.html)
SpeedVac SC110 Vacuum Concentrator Model SC110-120	Savant	n/a	Centrifugal vacuum concentrator for drying samples (under heat)
SDS Solution, 10% Sodium Dodecyl Sulfate Solution, Molecular Biology/ Electrophoresis	Fisher Scientific	BP2436200	
Sequencing Grade Modified Trypsin	Promega	V5111	
Sodium Chloride (Crystalline/ Certified ACS)	Fisher Scientific	S271-500	
TopTip, Empty, 10-200 µL, Pack of 96	Glygen Corporation	TT2EMT.96	Empty pipette tip with micron-sized hole used that can be used to pack chromatographic materials for enrichments, bundled with tube adapters

Triethylammonium bicarbonate buffer (TEAB, 1 M, pH 8.5 (volatile))	Sigma	90360-100ML	
Trifluoroacetic acid, Reagent Grade, 99%	Fisher Scientific	60-017-61	
Tris Base (White Crystals or Crystalline Powder/Molecular Biology)	Fisher Scientific	BP152-500	
Trypsin/Lys-C Mix, Mass Spec Grade	Promega	V5071	
Urea (Certified ACS)	Fisher Scientific	U15-500	
Water, Optima LC/MS Grade	Fisher Scientific	W64	