

Materials List for:

# Sample Preparation for Mass-spectrometry-based Proteomics Analysis of Ocular Microvessels

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## Materials

Name	Company	Catalog Number	Comments
<b>A. Chemicals</b>			
1, 4-Dithiothreitol (DTT)	Sigma-Aldrich	1.11474	
Calcium chloride dihydrate (CaCl <sub>2</sub> )	Carl Roth	5239.1	2.5 mM
Dulbecco's phosphate-buffered saline (PBS)	Thermo Fisher Scientific	14190169	
Formic acid (CH <sub>2</sub> O <sub>2</sub> )	AppliChem	A0748	
HPLC-grade acetonitrile (ACN, C <sub>2</sub> H <sub>3</sub> N)	AppliChem	A1605	
HPLC-grade methanol (CH <sub>3</sub> OH)	Fisher Scientific	M/4056/17	
HPLC-grade water	AppliChem	A1589	
Iodoacetamide (IAA)	Sigma-Aldrich	I6125	
Kalium chloride (KCl)	Carl Roth	6781.1	4.7 mM
Kalium dihydrogen phosphate (KH <sub>2</sub> PO <sub>4</sub> )	Carl Roth	3904.2	1.2 mM
LC-MS-grade acetic acid	Carl Roth	AE69.1	
Magnesium sulphate (MgSO <sub>4</sub> )	Carl Roth	261.2	1.2 mM
NuPAGE Antioxidant	Thermo Fisher Scientific (Invitrogen)	NP0005	
NuPAGE LDS Sample buffer	Thermo Fisher Scientific (Invitrogen)	NP0007	4x
NuPAGE MES SDS Running Buffer	Thermo Fisher Scientific (Invitrogen)	NP0002	20x
NuPAGE Sample reducing agent	Thermo Fisher Scientific (Invitrogen)	NP0004	10x
SeeBlue Plus2 pre-stained protein standard	Thermo Fisher Scientific (Invitrogen)	LC5925	
Sequencing grade modified trypsin	Promega	V5111	
Sodium chloride (NaCl)	Carl Roth	9265.2	118.3 mM
Sodium hydrogen carbonate (NaHCO <sub>3</sub> )	Carl Roth	965.3	25 mM
Trifluoroacetic acid (TFA, C <sub>2</sub> HF <sub>3</sub> O <sub>2</sub> )	Merck Millipore	108178	
α-(D)-(+)- Glucose monohydrate	Carl Roth	6780.1	11 mM
<b>B. Reagents and Kits</b>			
0.5mm zirconium oxide beads	Next Advance	ZROB05	
1.0mm zirconium oxide beads	Next Advance	ZROB10	

Colloidal Blue Staining Kit	Thermo Fisher Scientific (Invitrogen)	LC6025	To stain 25 mini gels per kit
NuPAGE 4-12 % Bis-Tri gels	Thermo Fisher Scientific (Invitrogen)	NP0321BOX	1.0 mm, 10-well
Pierce Bicinchoninic Acid (BCA) Protein Assay Kit	Thermo Fisher Scientific	23227	
ProteoExtract Transmembrane Protein Extraction Kit, TM-PEK	Merck Millipore	71772-3	20 reactions per kit
Tissue Protein Extraction Reagent (T-PER)	Thermo Scientific	78510	
<b>C. Tools</b>			
96-well V-bottom plates	Greiner Bio-One	651180	
Corning 96-well flat-bottom plates	Sigma-Aldrich	CLS3595-50EA	
Disposable microtome blades	pfm Medical	207500014	
Disposable scalpels #21	pfm Medical	200130021	
Dissection pins	Carl Roth	PK47.1	
Extra Fine Bonn Scissors	Fine Science Tools	14084-08	
Falcon conical centrifuge tubes (50 mL)	Fisher Scientific	14-432-22	
Mayo scissors, Tough cut	Fine Science Tools	14130-17	
Precision tweezers	Fine Science Tools	11251-10	Type 5
Precision tweezers, straight with extra fine tips	Carl Roth	LH53.1	Type 5
Self-adhesive sealing films for microplates	Ratiolab (vWR)	RATI6018412	
Standard pattern forceps	Fine Science Tools	11000-12	
Student Vannas spring scissors	Fine Science Tools	91501-09	
Vannas capsulotomy scissors	Geuder	19760	Straight, 77 mm
ZipTip <sub>C18</sub> pipette tips	Merck Millipore	ZTC18S096	
<b>D. Equipment and devices</b>			
150 × 0.5 mm BioBasic C18 column	Thermo Scientific, Rockford, USA	72105-150565	
30 × 0.5 mm BioBasic C18 pre-column	Thermo Scientific, Rockford, USA	72105-030515	
Amicon Ultra-0.5 3K Centrifugal Filter Devices	Merck Millipore	UFC500396	Pack of 96.
Analytical balance	Sartorius	H51	
Autosampler	CTC Analytics AG, Zwingen, Switzerland	HTS Pal	
BBY24M Bullet Blender Storm	Next Advance	NA-BB-25	
Eppendorf concentrator, model 5301	Sigma-Aldrich	Z368172	
Eppendorf microcentrifuge, model 5424	Fisher Scientific	05-403-93	Non-refrigerated
Heraeus Primo R Centrifuge	Thermo Scientific	75005440	Refrigerated
Labsonic M Ultrasonic homogenizer	Sartorius	BBI-8535027	
LC-MS pump, model Rheos Allegro	Thermo Scientific, Rockford, USA	22080	
LTQ Orbitrap XL mass spectrometer	Thermo Scientific, Bremen, Germany		

Multiskan Ascent plate reader	Thermo Labsystems	v2.6	
Rotator with vortex	neoLab	7-0045	
Titanium probe (Ø 0.5mm, 80mm long)	Sartorius	BBI-8535612	
Ultrasonic bath, type RK 31	Bandelin	329	
Xcell Surelock Mini Cell	Life Technologies	EI0001	