

MtoZ Biolabs Sample Submission Guidelines for Proteomics

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Introduction

In proteomics research, sample quality directly determines the depth, accuracy, and interpretability of downstream data. Whether for quantitative proteomics, post-translational modification (PTM) analysis, or protein interaction network studies, factors such as protein abundance, purity, integrity, and background interference in the sample significantly impact protein extraction efficiency, enzymatic digestion, and mass spectrometry sensitivity.

High-quality samples contribute to reproducible and statistically significant protein expression data, enhance detection of low-abundance proteins or modified peptides, and are essential for ensuring the reliability of scientific findings and their biological relevance. Conversely, improper sample handling can lead to protein degradation, loss of chemical modifications, or interference from contaminants, ultimately resulting in data deviation or project failure.

To ensure the accuracy and reproducibility of proteomics analysis, standardized sample preparation and submission procedures are critical. MtoZ Biolabs provides comprehensive proteomics solutions, including qualitative proteomics, quantitative proteomics, and PTM analysis, covering mainstream strategies such as DDA, DIA, TMT, and label-free methods. To assist clients in submitting samples smoothly, we have prepared this guide, which details requirements for sample types, collection, storage, and submission quantities.

Sample Types and Submission Requirements

1. Qualitative Proteomics

1.1 General Qualitative Proteomics

Sample Type	Submission Requirements
Gel spot/gel strip	Coomassie- or silver-stained gel spots or strips. Coomassie bands should be faintly visible to the naked eye, silver-stained bands should be clearly visible with no degradation. Submit either the target band or a 1-2cm gel lane. Place in a microcentrifuge tube with 0.5-1 mL ddH ₂ O to keep moist. Store at 4°C and ship with ice packs.
Protein solution	20µg of protein solution. Please inform us of buffer components in advance to avoid interference with downstream procedures. Ship on dry ice.
FFPE (paraffin section)	5-10µm thickness, 50mm ² area, more than 10 sections. Ship at room temperature.

1.2 Interaction-Based Qualitative Proteomics

Sample Type	Co-IP/Pull-down					
	MS Analysis		Interaction + MS		Interaction + MS + WB	
	Recommended quantity	Minimum quantity	Recommended quantity	Minimum quantity	Recommended quantity	Minimum quantity
Gel spot/ Gel strip	Target band or 1-2cm gel lane		/		/	
Agarose beads/ Magnetic beads	20-30 μ L		/		/	
Protein solution	10 μ g	5 μ g	1000 μ g	500 μ g	2000 μ g	1000 μ g
Tissue	Same as 2.1		Same as 2.2		Same as 2.3	

Note: Except for gel samples shipped with ice packs and FFPE samples shipped at room temperature, all other samples must be shipped on dry ice.

2. Quantitative Proteomics

2.1 General Quantitative Proteomics (Label-Free / DIA / TMT)

Sample Type		Recommended quantity	Minimum quantity
Gel spot/gel strip	Soft tissues (e.g., brain, heart, liver, spleen, lung, kidney, muscle, skin)	20mg	10mg
	Hard tissues (e.g., bone, cartilage, hair)	100mg	50mg
	Tumor tissue (tumor or adjacent connective tissue-containing samples)	30mg	15mg
Plant Tissue	Soft plant tissue (e.g., leaves and flowers of woody plants, grasses, algae, ferns, large fungi)	2g	1g
	Hard plant tissue (e.g., roots, bark, branches, seeds, fruits)	5g	2g
	Pollen	100mg	50mg
Cell Pellet	Adherent or suspension cells	1×10^7	2×10^6
Microbes	Common bacteria, fungal cells (cell pellets)	50mg or 50 μ L	20mg or 20 μ L
Body Fluids	Serum/plasma (without high-abundance protein depletion)	10 μ L	5 μ L
	Serum/plasma (with high-abundance protein depletion)	50 μ L	25 μ L
	Urine	1ml	0.5ml
	Cerebrospinal fluid	0.1ml	0.05ml
	Milk	0.5ml	0.2ml
	Saliva	0.2ml	0.1ml
	Tears	0.2ml	0.1ml
	Aqueous humor/vitreous humor	0.1ml	0.05ml

Sample Type		Recommended quantity	Minimum quantity
Body Fluids	Bronchoalveolar lavage fluid (BALF)	1ml	0.5ml
Culture Supernatant	Fermentation/cell culture supernatant	10ml	5ml
FFPE	Paraffin sections	20 slices	10 slices
	Formalin-fixed tissue	30mg	15mg
Protein Solution	User-extracted protein solution (composition must be provided in advance)	20μL	10μL

Note: 4D proteomics adds an ion mobility dimension at the mass spectrometry acquisition stage. Sample preparation is the same as for 3D proteomics, and the sample submission requirements remain unchanged.

2.2 Post-translational modifications Proteomics

Sample Type		Phosphorylation/ Glycosylation		Acetylation/ Ubiquitination/ SUMOylation/Lactylation/ Methylation	
		Recommended quantity	Minimum quantity	Recommended quantity	Minimum quantity
Animal Tissue	Soft tissues (e.g., brain, heart, liver, spleen, lung, kidney, muscle, skin)	100mg	50mg	200mg	100mg
	Hard tissues (e.g., bone, cartilage, hair)	5g	3g	10g	5g

Animal Tissue	Tumor tissue (tumor or adjacent connective tissue-containing samples)	100mg	50mg	200mg	100mg
Plant Tissue	Soft plant tissue (e.g., leaves and flowers of woody plants, grasses, algae, ferns, large fungi)	2g	1g	10g	5g
	Hard plant tissue (e.g., roots, bark, branches, seeds, fruits)	5g	2g	10g	5g
Cell Pellet	Adherent or suspension cells	2×10^7	1×10^7	1×10^8	5×10^7
Microbes	Common bacteria, fungal cells (cell pellets)	500mg or 500 μ L	250mg or 250 μ L	500 μ L	250 μ L
Body Fluids	Serum/plasma (without high-abundance protein depletion)	50 μ L	20 μ L	100 μ L	50 μ L
Protein Solution	User-extracted protein solution (composition must be provided in advance)	2mg	1mg	5mg	2mg

2.3 Microproteomics

Sample Type		Recommended quantity
Animal Tissue	Soft tissues (e.g., brain, heart, liver, spleen, lung, kidney, muscle, skin)	2mg
	Tumor tissue (tumor or adjacent connective tissue-containing samples)	10mg
Cell Pellet	Adherent or suspension cells	1000
Microbes	Common bacteria, fungal cells (cell pellets)	20µL
Body Fluids	Serum/plasma (without high-abundance protein depletion)	5µL
	Serum/plasma (with high-abundance protein depletion)	10µL
	Urine	500µL
	Cerebrospinal fluid	100µL
	Milk	200µL
	Saliva	200µL
	Tears	200µL
	Aqueous humor/vitreous humor	100µL
	Bronchoalveolar lavage fluid (BALF)	1mL
FFPE	Paraffin sections	10 slices
	Formalin-fixed tissue	10mg
Protein Solution	User-extracted protein solution (composition must be provided in advance)	10µg

Note: Electrophoresis results are not provided for microproteomics, and there is a certain risk of experimental failure.

3. Special Proteomics

3.1 Metaproteomics

Sample Type	Recommended quantity	Minimum quantity
Feces	5g	2g
Intestinal Contents	5g	2g
Soil/Mud/Sand	10g	5g
Fermentation Residues	10g	5g
Wastewater/Seawater	0.5L	0.3L

Note: The default acquisition mode for metaproteomics is DIA. A metagenomic database must be provided, or a combined metagenomics + metaproteomics project must be commissioned.

3.2 Exosomal Proteomics

Sample Type	Exosome Characterization				Exosome Omics		
	EM	Particle Size	Flow Cytometry (perindex)	WB (perindex)	Proteomics (LFQ)	Metabolomics	RNA Sequencing
Plasma/Serum	0.3mL	0.3mL	0.5mL	0.5mL	1mL	4mL	4mL
Cell Culture Supernatant	10mL	10mL	20mL	50mL	50mL	50mL	50mL
Urine	5mL	5mL	10mL	25mL	10mL	50mL	50mL
Cerebrospinal Fluid	2mL	2mL	4mL	4mL	10mL	15mL	15mL
Semen/Seminal Plasma	0.3mL	0.3mL	0.5mL	0.5mL	0.5mL	5mL	5mL
Animal Tissue	1g	1g	2g	2g	3g	6g	3g

Purified Exosomes	15µL	15µL	20µL	20µL	50µL	100µL	100µL
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Note: Exosome characterization can be customized based on the client's specific targets of interest. The required sample amount will be determined according to the selected analyses.

4. Peptidomics

Sample Type		Peptide Identification/ Peptidomics		Immuno-peptidomics	
		Recommended quantity	Minimum quantity	Recommended quantity	Minimum quantity
Animal Tissue	Soft tissues (e.g., brain, heart, liver, spleen, lung, kidney, muscle, skin)	50mg	25mg	200mg	50mg
	Hard tissues (e.g., bone, cartilage, hair)	100mg	50mg	2g	1g
	Tumor tissue (tumor or adjacent connective tissue-containing samples)	60mg	30mg	200mg	100mg
Plant Tissue	Soft plant tissue (e.g., leaves and flowers of woody plants, grasses, algae, ferns, large fungi)	1g	500mg	/	/
	Hard plant tissue (e.g., roots, bark, branches, seeds, fruits)	5g	2g	/	/

Plant Tissue	Pollen	200mg	100mg	/	/
Cell Pellet	Adherent or suspension cells	1×10^7	5×10^6	1×10^7	5×10^6
Microbes	Common bacteria, fungal cells (cell pellets)	100 μ L	50 μ L	/	/
Body Fluids	Serum/plasma	100 μ L	50 μ L	500 μ L	300 μ L
	Urine	2mL	1mL	/	/
Culture Supernatant	Fermentation/cell culture supernatant	10mL	5mL	/	/
Protein Solution	User-extracted protein solution (composition must be provided in advance)	2mg	1mg	5mg	2mg

Sample Collection, Processing, Storage, and Transportation

To ensure the scientific integrity and reproducibility of proteomics data, sample collection must follow the basic principles of representativeness, accuracy, consistency, and timeliness. Collection procedures should minimize physiological disturbances and operational bias, and sampling conditions should remain consistent across experimental and control groups in terms of timing, conditions, and processing. To prevent protein degradation or nonspecific modifications, it is recommended that samples be immediately snap-frozen or lysed after collection.

1. Sample Collection and Shipping Requirements

- (1) Maintain the required storage temperature throughout collection and shipping. For multi-omics analysis, it is advised to aliquot the sample during collection to avoid repeated freeze-thaw cycles.
- (2) Use ultrapure water for all steps involving water during sample collection. For procedures requiring organic solvents, reagents should be of chromatographic grade or higher.
- (3) Use high-quality centrifuge tubes for sample handling. After filling, seal the tubes securely with sufficient sealing film to prevent leakage or contamination (recommended brands: Eppendorf, Axygen, Corning, etc.).
- (4) To avoid issues with instrument or software recognition, sample names should be in alphabetic characters only. If symbols are necessary, use underscores "_" instead of special characters.
- (5) Label samples using permanent markers in multiple locations. Wrap tubes with shock-absorbing material to prevent damage during low-temperature transportation.
- (6) Use double-layered foam boxes for shipping. Include sufficient dry ice: 5 kg per day in summer and 4 kg per day in winter. Prepare the amount of dry ice based on expected shipping duration.

2. Sample Collection, Processing, Storage, and Transportation by Sample Type

2.1 Animal Tissue Samples

2.1.1 Regular Animal Tissues (brain, heart, liver, spleen, kidney, lung, etc.)

- (1) Accurately dissect the desired tissue. Immediately remove fat and connective tissues. Cut the sample into small pieces of 20–50 mg (roughly soybean-sized).
- (2) Rinse briefly in saline or PBS to remove blood and debris.
- (3) Transfer the sample into a pre-labeled centrifuge tube using tweezers, snap-freeze in liquid nitrogen for 10 minutes, then store at -80°C .
- (4) Ship on dry ice. Avoid freeze-thaw cycles.

Note: Tumor samples should be dissected according to pathological conditions. For adjacent tissues, stay at least 2 cm away from the lesion.

2.1.2 Hard Animal Tissues (bone, cartilage, etc.)

- (1) Wash in PBS. Cut cartilage into $\sim 1\text{ cm}^3$ cubes with a scalpel.
- (2) Transfer to labeled centrifuge tube, snap-freeze in liquid nitrogen for 10 minutes, store at -80°C .
- (3) Ship on dry ice. Avoid freeze-thaw cycles.

2.1.3 Hard Tissues (Hair)

- (1) Rinse the sample with 2% SDS in 50 mM sodium phosphate buffer (pH 7.8) to remove contaminants.
- (2) Air-dry and transfer to a labeled centrifuge tube, store at -80°C .
- (3) Ship on dry ice. Avoid freeze-thaw cycles.

2.1.4 Soft-Bodied Organisms (e.g., schistosomes, trichinella)

- (1) Collect the organism and rinse thoroughly with pre-chilled PBS to remove host material.
- (2) Transfer to a labeled centrifuge tube, snap-freeze in liquid nitrogen for 10 minutes, store at -80°C .
- (3) Ship on dry ice. Avoid freeze-thaw cycles.

2.2 Plant Tissue Samples

2.2.1 Regular Plant Tissues (leaves, flowers, fruits, seeds, roots, bark, ferns, etc.)

- (1) Isolate target tissue, remove unwanted parts, rinse off dust and debris, and blot dry with lint-free paper.
- (2) Wrap in foil or place in labeled centrifuge tube, snap-freeze in liquid nitrogen for 10 minutes, store at -80°C .
- (3) Ship on dry ice. Avoid freeze-thaw cycles.

2.2.2 Pollen

- (1) Collect pollen during flowering using a dissecting microscope. Remove anther fragments using dissection needles and transfer into an Eppendorf tube.
- (2) Check morphology and purity under light microscopy. Store at -80°C .
- (3) Ship on dry ice. Avoid freeze-thaw cycles.

2.2.3 Algae

- (1) Use appropriate centrifugation speed for different algae species to preserve integrity. Wash with PBS 2-3 times. Snap-freeze for 10 minutes in liquid nitrogen, then store at -80°C .
- (2) Ship on dry ice. Avoid freeze-thaw cycles.

2.3 Cell Samples

2.3.1 Suspension Cells

- (1) Centrifuge at 1,000 g for 10 minutes to collect cells; discard supernatant.
- (2) Wash with cold PBS 2-3 times. Centrifuge and discard the supernatant. Collect pellet into a 1.5 mL centrifuge tube.
- (3) Snap-freeze in liquid nitrogen for 10 minutes. Store at -80°C .
- (4) Ship on dry ice. Avoid freeze-thaw cycles.

2.3.2 Adherent Cells

- (1) Discard media, invert dish onto absorbent paper to remove liquid.
- (2) Add cold PBS, rock gently for 1 minute to rinse. Discard PBS.
(Repeat steps (1) and (2) twice to discard the culture medium)
- (3) Digest cells with trypsin on ice. Resuspend in cold PBS.
- (4) Collect cell suspension into 15 mL tubes, centrifuge at 1,000 g for 10 minutes at 4°C . Discard PBS.
- (5) Resuspend in 1 mL PBS, transfer to 1.5 mL tubes, centrifuge, discard PBS, and retain pellet. Snap-freeze 10 minutes in liquid nitrogen, store at -80°C .
- (6) Ship on dry ice. Avoid freeze-thaw cycles.

2.4 Liquid Samples

2.4.1 Serum

- (1) Collect blood using vacuum tubes without anticoagulant (red cap recommended) or clean tubes.
- (2) Gently invert 8-10 times, rest at 4°C for 30 minutes.
- (3) Centrifuge at 1,000 g for 10 minutes at 4°C .
- (4) Transfer yellow supernatant (serum) to clean tube, mix, and spin briefly.

- (5) Aliquot 100–200 µL per tube. Snap-freeze in liquid nitrogen for 10 minutes, store at –80°C.
- (6) Ship on dry ice. Avoid freeze-thaw cycles.

2.4.2 Plasma

- (1) Collect blood in anticoagulant tubes (EDTA, purple cap recommended).
- (2) Invert gently 8–10 times.
- (3) Centrifuge at 1,000 g for 10 minutes at 4°C.
- (4) Transfer plasma to clean tube, mix, and spin briefly.
- (5) Aliquot 100–200 µL per tube. Snap-freeze in liquid nitrogen, store at –80°C.
- (6) Ship on dry ice. Avoid freeze-thaw cycles.

Note: Handle gently to avoid hemolysis. Samples should appear clear yellow; pink or red indicates hemolysis and may impact results.

2.4.3 Urine

- (1) Collect midstream first-morning urine. Store temporarily at 4°C (not exceeding 8 hours). Avoid bacterial contamination.
- (2) Centrifuge at 1,000 g for 15 minutes at 4°C to remove debris.
- (3) Transfer supernatant to new tube, aliquot 1 mL per tube. Snap-freeze in liquid nitrogen, store at –80°C.
- (4) Ship on dry ice. Avoid freeze-thaw cycles.

2.4.4 Milk / Ascites Fluid

- (1) Collect using clinical procedures into centrifuge tubes.
- (2) Aliquot 1 mL per tube as needed. Snap-freeze 10 minutes in liquid nitrogen, store at –80°C.
- (3) Ship on dry ice. Avoid freeze-thaw cycles.

2.4.5 CSF, Lymph, Synovial, and Other Puncture Fluids

- (1) Collect using clinical procedures.
- (2) Centrifuge at 1,000–2,000 g for 5 minutes at 4°C.
- (3) (Optional) Filter supernatant with 0.22 µm membrane.
- (4) Aliquot 100 µL per tube, snap-freeze, store at –80°C.

2.4.6 Saliva

- (1) Fast at least 2 hours. Collect between 9 a.m. and 12 p.m.
- (2) Centrifuge at 1,000–2,000 g for 5 minutes at 4°C.
- (3) (Optional) Filter with 0.22µm membrane.
- (4) Aliquot 100–200 µL per tube. Snap-freeze, store at –80°C.
- (5) Ship on dry ice. Avoid freeze-thaw cycles.

2.4.7 Tears

- (1) Use capillary pipettes for collection. Centrifuge at 8,000–14,000 g for 5 minutes at 4°C.
- (2) Transfer supernatant to tubes, snap-freeze, store at –80°C.
- (3) Ship on dry ice. Avoid freeze-thaw cycles.

2.4.8 Culture Medium Supernatant

- (1) Remove media, collect cells following cell protocols.
- (2) Replace with serum-free medium and continue culture for 24–48 hours based on growth rate.
- (3) Centrifuge at 300 g for 10 minutes at 4°C.
- (4) Transfer supernatant to a 15mL tube, centrifuge again at 3,000g for 15 minutes to remove debris.
- (5) Transfer clarified supernatant to clean tubes, aliquot 5–10mL per tube. Snap-freeze, store at –80°C.
- (6) Ship on dry ice. Avoid freeze-thaw cycles.

Note: Serum-containing media introduces high-abundance proteins, reducing protein identification.

2.5 Microorganisms

- (1) Centrifuge at 5,000 g for 15 minutes at 4°C, discard supernatant.
- (2) Wash pellet with cold PBS, centrifuge and discard supernatant. Repeat 3 times.
- (3) Transfer pellet to clean tube, aliquot 50–100 µL per tube. Snap-freeze, store at –80°C.
- (4) Ship on dry ice. Avoid freeze-thaw cycles.

For sample types not covered in this guideline, or if you have any questions regarding sample preparation, processing, packaging, or shipping, please feel free to contact us. MtoZ Biolabs has an experienced proteomics technical support team ready to provide one-on-one consultation and sample preparation guidance. We are committed to helping you deliver your samples in optimal condition, ensuring reliable data and project success.