

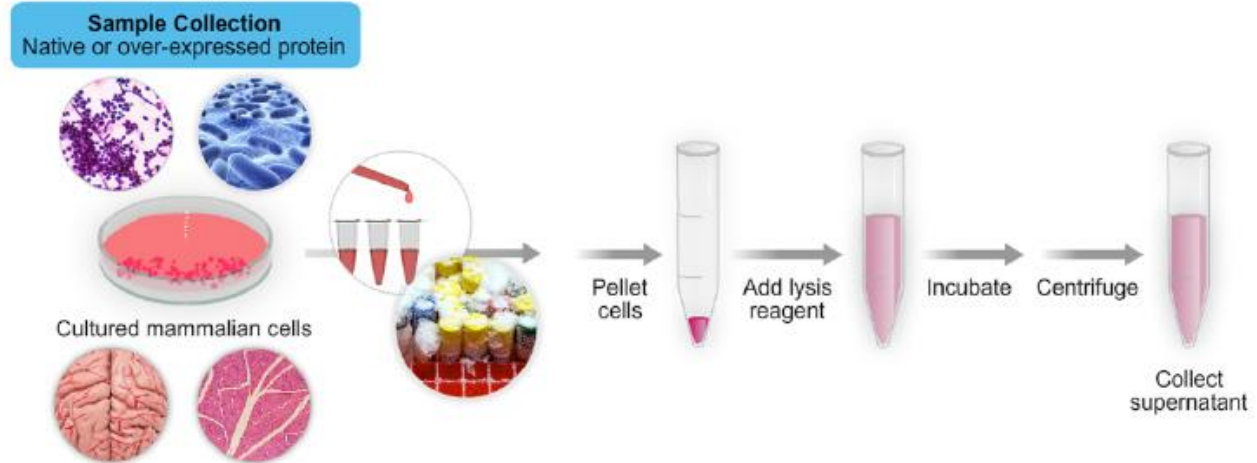
Overview of Protein Sample Preparation

Steps for obtaining proteins:

- Sample collection
- Cell lysis
- Protein extraction and stabilization

Proteins typically extracted from:

- Cultured mammalian cells
- Mammalian tissues
- Primary cells



Lysis Buffer composition:

Purpose

50 mM Tris-HCl (pH 8.0)	buffer salt
150 mM NaCl	maintain ionic strength of medium
1 mM EDTA	reduce oxidation damage, chelate metal ions
100 mM NaF	serine/threonine phosphatase inhibitor
1 mM MgCl ₂	stabilization
10% Glycerol	stabilization of proteins
1% Triton	solubilization of poorly soluble proteins
V = 200 mL	

1 µg/ml leupeptin (cysteine and serine protease inhibitor) or 0.1-1.0 mM PMSF (Protease/phosphatase inhibitor). PMSF has a short half-life time in aqueous solutions. A stock solution of 100 mM in isopropanol should be made and diluted into buffer immediately before use.