

## RIPA Lysis Buffers – Instruction Manual

### Catalogue Information

Catalogue No.	Product Name	Pack Size
BL504A	RIPA Lysis Buffer - Strong	100 mL
BL651A	RIPA Lysis Buffer - Medium	100 mL
BL652A	RIPA Lysis Buffer - Mild	100 mL

### Product Description

RIPA (Radio Immunoprecipitation Assay) Lysis Buffers are classical formulations designed for rapid and efficient cell and tissue lysis, allowing extraction of membrane, cytoplasmic, and nuclear proteins. The buffers are available in three strengths – Strong, Medium, and Mild – to accommodate different sample types and experimental requirements. Protein lysates obtained with RIPA buffers are suitable for Western blotting, immunoprecipitation (IP), and related applications. Due to the presence of detergents, protein quantification should be performed using the BCA method, not the Bradford assay.

### Composition Comparison

Buffer Type	Tris (pH 7.4) / NaCl	Detergents	Additives	Key Features
Strong (BL504A)	50 mM / 150 mM	1% Triton X-100, 1% Sodium deoxycholate, 0.1% SDS	Sodium orthovanadate, sodium fluoride, EDTA, leupeptin, PMSF (add fresh)	High-strength lysis; suitable for membrane, nuclear, and phosphorylated proteins
Medium (BL651A)	50 mM / 150 mM	1% NP-40, 0.5% Sodium deoxycholate, 0.1% SDS	Sodium orthovanadate, sodium fluoride, EDTA, PMSF (add fresh)	Balanced lysis; preserves protein-protein interactions; suitable for phosphoproteins
Mild (BL652A)	50 mM / 150 mM	1% NP-40, 0.25% Sodium deoxycholate	Sodium orthovanadate, sodium fluoride, EDTA, PMSF (add fresh)	Gentle lysis; ideal for cytoplasmic proteins and co-immunoprecipitation



RIPA buffer strength and detergent composition determine their lysis capability and protein compatibility. Strong buffer provides the most aggressive lysis for membrane and nuclear proteins; medium offers balance; mild is optimized for preserving interactions and gentle lysis applications.

#### **Instructions for Use**

1. Before use, thaw the RIPA buffer completely and mix well. Add Protease Inhibitor (WSBL630A) and/or Phosphatase Inhibitor (WSBL636A) just prior to use. Keep all reagents and samples on ice or at 4 °C during lysis.
2. For adherent cells: Remove culture medium, wash once with PBS or serum-free medium if required, and add 150–250 µL buffer per well of a 6-well plate. Pipette several times to aid lysis. Cells should lyse within 1–2 seconds.
3. For suspension cells: Collect cells by centrifugation and resuspend pellet. Add 150–250 µL buffer per 0.5–1×10<sup>6</sup> cells. Mix or flick gently to lyse completely. Avoid visible cell clumps.
4. For tissue samples: Mince tissue into small pieces. Add 150–250 µL buffer per 20 mg tissue. Homogenize using a glass homogenizer until fully lysed. Centrifuge at 10,000–14,000 × g for 3–5 minutes and collect supernatant for downstream applications.

#### **Notes**

1. Lysates may contain a small clear gel-like mass composed of genomic DNA. This is normal. If tight DNA-protein complexes need to be analyzed, sonicate before centrifugation.
2. Avoid repeated freeze–thaw cycles; aliquot if necessary.
3. Protease inhibitors should be added freshly before use.
4. Suitable for BCA protein assay; not compatible with Bradford assay.
5. Always wear a lab coat and gloves during handling.

#### **Storage and Stability**

1. Store at –20 °C
2. Valid for 1 year when stored under recommended conditions
3. After addition of protease inhibitors, aliquot and freeze to avoid repeated freeze–thaw cycles

#### **Notice**

For in vitro research use only, not for diagnostic or therapeutic use. This product is not a medical device.

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