ALPHA DIAGNOSTIC INTERNATIONAL

Product Specification Sheet

RIPA buffers (with or without protease and phosphatase inhibitors)

½ **Cat.** # RIPA-50	RIPA lysis buffer kit for mammalian cell lysis buffer (complete with protease phosphatase inhibitors cocktail)	**SIZE**: 50 ml
½ **Cat.** # RIPA-51	RIPA lysis buffer kit for mammalian cell lysis buffer (**without** protease and phosphatase inhibitor cocktail ate)	**SIZE**: 50 ml

Most researchers study expression or changes in a given protein in various cultured cells or tissues from animal or human origin. The first step is to harvest the proteins by disrupting the cells (lysis) and homogenization and extraction of cellular proteins. Many methodologies including homogenization, centrifugation and sonication have been employed to prepare cell or tissue extracts for further analyses by ELISA, Western, and IP etc. Many proteins are susceptible to rapid proteolysis or fragmentation of proteins after cell lysis or tissue disruption. Therefore, it is very important to minimize proteolysis and keep the phosphatase inactive.

RIPA (**R**adio-**I**mmuno**p**recipitation **A**ssay) Lysis Buffer is the most common buffer for rapid, efficient cell lysis and solubilization of proteins from both adherent and suspension cultured mammalian cells. It is widely used for cell lysis followed by immunoprecipitation (IP or co-IP) or direct western blotting. Most antibodies and protein antigens are not adversely affected by the components of this buffer. In addition, RIPA Lysis Buffer minimizes nonspecific protein-binding interactions to keep background low, while allowing most specific interactions to occur, enabling studies of relevant protein-protein interactions.

The following RIPA Lysis Buffer components have the following effects:

- Tris-HCl is a buffering agent prevents protein denaturation
- NaCl is a salt that prevents non-specific protein aggregation
- NP-40 is a non-ionic detergent to extract proteins
- Na-deoxycholate and SDS are ionic detergents to extract proteins
- sodium azide is a bacteriostatic agent added to retard bacterial growth.
- Protease inhibitors (PMSF, Leupetin, pepstatin, AEBSF, aprotinin, bestatin, E-64) added broad spectrum protease inhibitor cocktail with proprietary formulation.
- Phosphatase inhibitors (EDTA, sodium fluoride, and orthovanadate)

RIPA Lysis Buffer is supplied as convenient and ready-to-use solution that requires no preparation. It is available with or without Protease and phosphatase inhibitors. Unless the specified reagents interfere with a given protein function or analyses, it is advisable to use the RIPA formulation with the protease and phosphatase inhibitors.

Ready to use RIPA buffer not only saves time but it avoids the hassle of buying many different reagents from various suppliers and it is also more cost effective.

Buffer Composition

RIPA Buffer (cat #RIPA-51)

1X RIPA lysis buffer consists of 50 mM Tris HCl, 150 mM NaCl, 1.0% (v/v) NP-40, 0.5% (w/v), Sodium Deoxycholate, 1.0 mM EDTA, 0.1% (w/v) SDS and 0.01% (w/v) sodium azide at a pH of 7.4.. Store at 4oC or at -20oC for long-term in suitable size aliquots.

RIPA Buffer (cat #RIPA-50)

RIPA-51 plus a proprietary cocktail of protease inhibitors consisting of PMSF, Leupetin, pepstatin, AEBSF, aprotinin, bestatin, E-64) and Phosphatase inhibitors (EDTA, sodium fluoride, and orthovanadate). Store at 4oC or at -20oC for long-term in suitable size aliquots.

Supplied as 2-compnent kit.

1. RIPA buffer (#RIPA-51), 50 ml
2. Protease inhibitors 100x (0.5ml)

Note: PMSF, a broad spectrum, protease inhibitor can be added if necessary. It is supplied as 100X stock. Add 10 ul/ml or 0.5 ml/50 ml stock. Add just before use. Store at 4oC.

Form & Storage

Storage
Store at 4oC for at least 6 months.

Stability: 6-12 months at 4oC.

Shipping: 4oC for solutions and room temp for powder.

General References: (1). Harlow E and Lane D (1999) Using antibodies, A laboratory manual, Cold Spring Harbor Press.

*This product is for in vitro research use only.

RIPA50-51-RIPA-Cell-Tissue-Lysis-buffer 150508A

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