EURx® MOLECULAR BIOLOGY PRODUCTS

Mild Cell Lysis

Cat. No.	size
E0292-01	50 ml
E0292-02	200 ml

Storage and transport: 4°C.

Term of validity: 12 months

Content: 50 mM Tris-HCl pH 7.6, 250 mM NaCl, 1 % detergent, 5 mM EDTA, 0.02 % NaN_3

Product Important Information: Mild Cell Lysis does not contain protease inhibitors (1 mM PMSF or Protease Inhibitor Cocktail), which should be added directly before proteins extraction.

For extraction of nucleic and mitochondrial proteins it is recommended to use RIPA buffer (E0295).

Mild Cell Lysis is a gentle lysis solution reagent used for efficient total lysis of proteins from adherent and suspension cultured mammalian cells and tissues, depending on nonionic detergent and recommended for cytoplasmic and membrane-bound proteins extraction. Extracted proteins can be used for PAGE analysis, Western blotting, IP, ELISA.

Procedure for Lysis of Monolayer-cultured Adherent Mammalian Cells:

• Carefully remove culture medium from adherent cells.

• Wash cells twice with chilled PBS.

• Add cold Mild Cell Lysis to the cells (1 mL of buffer per 10^8 cells - the volume might need optimization for different cell types and amount of the protein of interest).

Incubate on ice for 15 minutes, swirling the plate occasionally until the clear lysate is visible.

• Collect the lysate and centrifuge at ~14,000 × g for 15 minutes.

• Transfer supernatant to a new tube for further analysis or store at -80 °C.

Procedure for Lysis of Suspension-cultured Mammalian Cells:

• Pellet the cells by centrifugation at 2500 × g for 5 minutes. Carefully remove and discard the supernatant.

• Wash cells twice in chilled PBS. Centrifuge at 2500 × g for 5 minutes. Carefully remove and discard the supernatant.

• Add cold Mild Cell Lysis to the cell pellet (1 mL of buffer per 10^8 cells - the volume might need optimization for different cell types and amount of the protein of interest). Pipette the mixture up and down to suspend the pellet.

• Shake mixture gently for 15 minutes on ice until the clear lysate is visible.

• Centrifuge samples at 14,000 × g for 15 minutes.

• Transfer supernatant to a new tube for further analysis or store at -80 °C.

Procedure for Lysis of Tissues:

• Place the fresh tissue into chilled PBS and rinse several times. Mince the tissue into small pieces.

• Add cold Mild Cell Lysis to the tissue at 10:1 (10 mL Mild Cell Lysis per gram of tissue).

• Homogenize for several minutes at high speed until no tissue chunks remain.

• Incubate on ice for 3 minutes.

• Centrifuge at 10000 x g for 10 minutes

• Transfer supernatant to a new tube for further analysis or store at -80 °C.

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