

## MDA-MD-435S Whole Cell Lysate

**Cat. No.** MDA-MD-435S-20H    **Lot. No.** (See product label)

### SPECIFICATION

**Species** Human

**Concentration** 2 mg/ml

**Tissue Type** mammary gland ductal carcinoma

**Preparation method**

The cells were grown in Leibovitz's L-15 medium with 0.01 mg/ml insulin, supplemented with 10% fetal bovine serum. Cells were washed with PBS and then incubated on ice in modified RIPA buffer, containing 150 mM sodium chloride, 50 mM Tris HCl, pH 7.4, 1 mM EDTA, 1.0% NP-40, 0.5% sodium deoxycholic acid and 0.1% SDS, to lyse the cells. Protein integrity was ensured using a cocktail of protease inhibitors with broad specificity for the inhibition of aspartic, cysteine, and serine proteases as well as aminopeptidases (0.1 mM AEBSF HCl, 0.08 M Aprotinin, 5 M Bestatin, 1.5 M E-64, 2 M Leupeptin Hemisulfate, 1 M Pepstatin A). Phosphatase inhibitors 1 mM NaF and 1 mM Na<sub>3</sub>VO<sub>4</sub> were also added. Cell debris was removed by centrifugation. Protein concentration was determined by a modified Lowry assay using a commercially available kit. Protein concentration was adjusted to 2 mg/ml and then an equal volume of 2X SDS-PAGE sample buffer was added.

**Recommended Usage**

For research use only, not for diagnostic or therapeutic use.

**Storage Buffer**

1X SDS-PAGE Sample Buffer (62.5 mM Tris HCl, 2% SDS, 10% Glycerol and 0.005% bromophenol blue, pH 6.8)

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### Applications

Protein Lysate for WB. Ready-to-use lysates are especially prepared as positive controls for separation by SDS-PAGE and subsequent western blot analysis. Lysates are prepared in denaturing buffer WITHOUT dissociating agents (i.e. no 2-mercaptoethanol or dithiothreitol has been added).

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