

Lysate Preparation

Calyculin A is a serine/threonine phosphatase inhibitor that inhibits the activity of protein phosphatases PP1 and PP2A. Human carcinoma A431 cells treated with calyculin A for 30 minutes can undergo significant threonine phosphorylation, as shown by western blotting using anti-Phospho-Akt (Thr-34), cat.# AP1001, as compared to untreated, control cell lysates.

Confluent cultures of A431 cells were serum starved overnight. Cells were then either left untreated (Cat.# AL9001) or treated with Calyculin A at a final concentration of 100 nM for 30 minutes at 37°C (Cat. #AL9101). Cells were lysed in 1% SDS, 1.0 mM sodium ortho-vanadate, 1 mM sodium fluoride, 10 mM Tris (pH 7.4) buffer. Protein concentration was determined using the BCA method (Pierce) before diluting to final concentration and buffer.

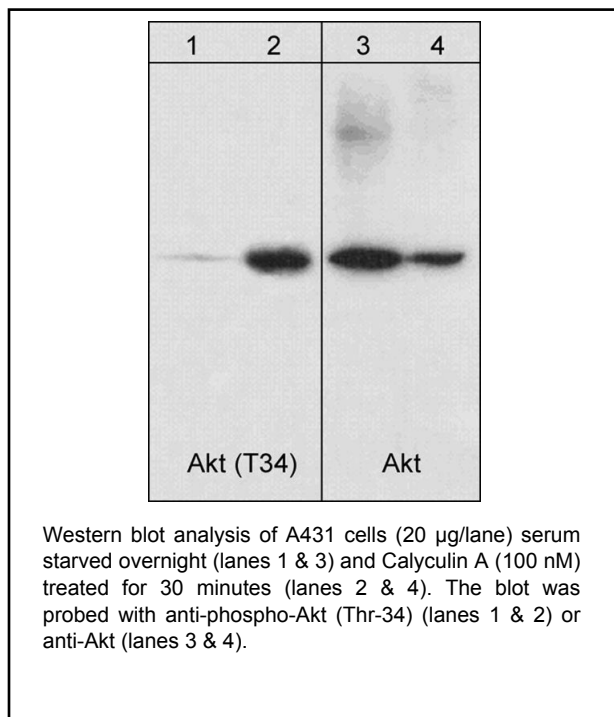
Buffer and Storage

Cell Lysates are supplied at a concentration of 1 mg/ml in electrophoresis sample buffer (62.5 mM Tris pH 6.8, 2% SDS, 5% glycerol, 0.003% bromophenol blue, 0.9% β-mercaptoethanol). Store at -20°C. Do not boil or dilute. Stable for 1 year.

Applications

WB 20 µl/lane

End user should determine optimal quantity for their particular applications and experiments.



Related Products

AL9101 A431 + Calyculin A (30min) Lysate

AL9201 A431 EGF Control Lysate

AL9301 A431 + EGF (5 min) Lysate

AL9401 A431 Pervanadate Control Lysate

AL9501 A431 + Pervanadate Lysate

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