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Product Datasheet

MCF-7 Cell Nuclear Extract (orb348687)

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Description ^{nts.}	MCF-7 Cell Nuclear Extract
Conjugation	Unconjugated
Tested Applications	WB
Preservatives	Preservative: None. Stabilizer: 10% (v/v) Glycerol. 1X SDS-PAGE Sample Buffer (62.5 mM Tris HCl, 2% SDS, 10% Glycerol and 0.005% bromophenol blue, pH 6.8)
Form/Appearance	Liquid (sterile filtered)
Concentration	1.0 mg/ml
Storage	Store vial at -70° C or COLDER. For extended storage, aliquot contents to minimize freeze/thaw cycles.
Note	For research use only
Application notes	ready-to-use nuclear extracts are especially prepared as positive controls for separation by SDS-PAGE and subsequent western blot analysis. Nuclear extracts are supplied in denaturing buffer without dissociating agents. Heat nuclear extract to 95°C for 5 minutes and rapidly cool. If dissociating conditions are desired add reducing agent prior to heating. The recommended loading volume per lane is 10-30 I depending on the size format of your gel.
Purity	The cells were grown in EMEM supplemented with 10% FBS (Fetal Bovine Serum). Cells were washed in PBS and incubated on ice in modified RIPA buffer containing 150 mM sodium chloride, 50 mM Tris Cl, 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 is 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 µM Aprotinin, 5 µM Bestatin, 1.5 µM E-64, 2 µM Leupeptin Hemisulfate and 1 µM Pepstatin A). Phosphatase inhibitors sodium fluoride, sodium orthovanadate, sodium pyrophosphate and [-glycerophosphate were also added. Cell debris was removed by centrifugation. Protein concentration was

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