

Chaps Cell Extract Buffer

Supplied as a 10X stock



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For Research Use Only. Not For Use In Diagnostic Procedures.

Product Includes	Product #	Quantity
Chaps Cell Extract Buffer (10X)	61536S	5 ml
30X Reducing Agent (1.25 M DTT)	14265P	0.25 ml

Description: Chaps Cell Extract Buffer can be used to lyse cells under nondenaturing conditions and is recommended for the preparation of cytoplasmic cell lysates to be used with our caspase signaling pathway antibodies (see Companion Products).

Solutions and Reagents:

10X CHAPS Cell Extract Buffer (#61536) 1X concentration:
50 mM Pipes/HCl (pH 6.5), 2 mM EDTA, 0.1% Chaps, 20 µg/ml Leupeptin, 10 µg/ml Pepstatin A, 10 µg/ml Aprotinin. Add DTT to 5mM.

30X Reducing Agent (1.25 M DTT) (#14265) 1X concentration:
5 mM DTT

Preparation of 1X Cell Extract Buffer:

- Buffer should be prepared immediately before use.
- Immediately before dilution, invert the 10X Chaps Cell Extract Buffer several times to suspend all buffer components.
- Add 1/10 volume of 10X Chaps Cell Extract Buffer to 9/10 volume of Milli-Q water or equivalently purified water.
- Add DTT to final concentration of 5 mM (1:250 dilution) and PMSF* to final concentration of 1 mM.

*reagent not included

Sample Preparation Using Chaps Cell Extract Buffer:

- Treat cells by adding fresh media containing regulator for desired time.
- Aspirate media from cultures; wash cells three times with PBS; aspirate. Scrape cells into PBS and spin down to pellet.
- Lyse cells by adding ice cold 1X Chaps Cell Extract Buffer (1 volume of cell pellet). Resuspend cells in the buffer, freeze at -80°C and thaw twice, centrifuge the lysate cold at 14,000 rpm. Keep the supernatant and discard the pelleted cell debris.
- Add SDS Sample Buffer and heat sample to 95–100°C for 5 minutes; cool on ice.
- Microcentrifuge for 5 minutes.
- Load 5 µL onto SDS-PAGE gel (10 cm x 10 cm).

Storage: Store Chaps Cell Extract Buffer and 30X Reducing Agent (1.25 M DTT) at -20°C.

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.