



Description

CytoBuster Protein Extraction Reagent	50 ml	71009-3
	250 ml	71009-4

CytoBuster Protein Extraction Reagent is a proprietary formulation of detergents optimized for efficient extraction of soluble proteins from mammalian cells. The unique composition of CytoBuster enables isolation of functionally active proteins without secondary treatment such as sonication or freeze/thaw. CytoBuster Protein Extraction Reagent has been specifically formulated for use with Western blotting protocols, immunoprecipitation, and kinase/phosphatase assays. The buffer is compatible with protease inhibitors, kinase and phosphatase inhibitors, and with BCA protein assays.

Storage

CytoBuster should be stored at room temperature.

Using CytoBuster Protein Extraction Reagent

CytoBuster can be used directly for protein extraction at room temperature. If proteolysis must be minimized, CytoBuster may be chilled to 4°C and the extraction performed on ice. The addition of protease inhibitor cocktails (e.g. Cat. Nos. 539132 or 539134) may also be helpful.

Table 1: Recommended volumes of CytoBuster for extraction

Culture Format	Surface Area (cm ²)	Volume of CytoBuster
96 Well Plate	0.32	30 µl
48 Well Plate	0.8	50 µl
24 Well Plate	2.0	100 µl
12 Well Plate	4.0	200 µl
6 Well Plate	9.6	300 µl
35 mm Dish	9.6	300 µl
60 mm Dish	21.0	500 µl
100 mm Dish	55.0	1.0 ml
T-25 Flask	25.0	500 µl
T-75 Flask	75.0	1.5 ml
Suspension cells	10 ⁶ cells*	150 µl

*Suspension cells vary greatly in cell size; thus adjustment may be necessary.

Extraction of monolayer cells

- Aspirate culture medium from cells.
Optional: If components of the culture media (i.e. phenol red) are inhibitory to protein analysis, wash cells once with PBS (PBS; 43 mM Na₂HPO₄, 15 mM KH₂PO₄, 137 mM NaCl, 27 mM KCl, pH 7.4) or Hanks' Buffered Salts Solution (HBSS) prior to CytoBuster addition.
- Add the recommended amount of CytoBuster (from Table 1) and allow to extract at room temperature for 5 min.
- Scrape cell debris using a cell scraper or rubber policeman. Orient the plate so that all debris is pooled in the CytoBuster Protein Extraction Reagent.
- Remove extract to a suitably sized tube and centrifuge for 5 min at 16,000 × g (4°C).

Note: Longer centrifugation at higher speeds may be desired for certain applications.

- Remove supernatant to a new tube and proceed with analysis.

Note: Extracts prepared with CytoBuster can be used immediately or may be stored for extended periods of time at -20°C or -80°C.



CytoBuster™ Protein Extraction Reagent

Extraction of suspension cells

1. Pellet the cells by low speed centrifugation (e.g 5 min at 2500 × g).
Optional: If components of the culture media (i.e. phenol red) are inhibitory to reporter enzyme analysis, wash cells once with PBS or HBSS prior to CytoBuster addition. Pellet cells as above and discard supernatant.
2. Resuspend the cells in CytoBuster using 150 µl per 10⁶ cells (the optimal amount to use may vary based on cell size).
3. Incubate at room temperature for 5 min.
4. Remove extract to a suitably sized tube and centrifuge for 5 min at 15,000 × g (4°C).
5. Transfer supernatant to a fresh tube and proceed with analysis.

Related products

Product	Size	Cat. No.
Reportasol™ Extraction Buffer	25 ml	70909-3
	125 ml	70909-4
Reporter Assay	500 assays	70979-3
BetaFluor™ β-Galactosidase Assay Kit	2500 assays	70979-4
BetaRed™ β-Galactosidase Assay Kit	500 assays	70978-3
	2500 assays	70978-4
FRETWorks™ S•Tag Assay Kit	100 assays	70724-3
	1000 assays	70724-4
S•Tag™ Rapid Assay Kit	100 assays	69212-3
pTriEx Multisystem Expression Vectors	see catalog	
Transfection and DNA Purification		
GeneJuice™ Transfection Reagent	1 ml	70967-3
	10 ml	70967-4
Mobius™ 200 Plasmid Kit	25 rxn	70970-3
Mobius 200 Columns	25 rxn	71019-3
Mobius 500 pET Plasmid Kit	10 rxn	70969-3
Introductory Mobius 1000 Plasmid Kit	2 rxn	70854-3
Mobius 1000 Plasmid Kit	10 rxn	70853-3
	25 rxn	70853-4
Introductory UltraMobius™ 1000 Plasmid Kit	2 rxn	70907-3
UltraMobius 1000 Plasmid Kit	10 rxn	70906-3
	25 rxn	70906-4
ClearSpin™ Filter	pkg/25	70848-3
Mobius Buffer Kit	10 rxn	70855-3
Mobius 1000 Columns	10 rxn	70849-3
	25 rxn	70849-4