# invitrogen

# Cell Lysis Buffer (20 mL)

Cat no.	Volume
EPX-99999-000	20 mL

**Description:** Cell Lysis Buffer is used to lyse cells under nondenaturing conditions.

# **Cell Lysis Buffer**

- 20 mM Tris-HCl (pH 7.5)
- 150 mM NaCl
- 1 mM Na<sub>2</sub>EDTA
- 1 mM EGTA
- 1% Triton
- 2.5 mM sodium pyrophosphate
- 1 mM β-glycerophosphate
- 1 mM Na<sub>3</sub>VO<sub>4</sub>
- 1 μg/ml leupeptin
- 1. If buffer will be continually used, it is recommended that the cell lysis buffer be kept at 4°C for 1-2 weeks. For longer periods of time, buffer should be stored at -20°C. Aliquotting of cell lysis buffer is recommended if many small experiments are to be performed.
- 2. Thaw cell lysis buffer at 24-30°C, mixing end-over-end.
- 3. Chill cell lysis buffer on ice and add PMSF just prior to use.

Note: CST recommends adding 1 mM PMSF immediately before use

### Lysis

For lysis of adherent cells, we recommend the following: (all reagents and lysates must be kept cold)

- Treat cells as desired.
- 2. Wash plate with PBS to remove residual media.
- 3. Add  $400 \mu L$  of cell lysis buffer / 10 cm dish.
- 4. Incubate plate on ice for 5 min.
- Scrape cells.
- 6. Sonicate briefly.
- 7. Spin extract 10 min at 14,000 x g in a cold microfuge.
- 8. Remove supernatant for use.

#### **Additional Notes:**

- 1. For non-adherent cells, add 400 µl of buffer per 10<sup>7</sup> cells once they have been washed in 1X PBS and pelleted.
- 2. Additional protease inhibitors can be added to the cell lysis buffer without any difficulties.

**Storage:** Store at -20°C. For short term storage (1- 2 weeks), cell lysis buffer can be stored at 4°C.



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Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

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