

## HeLa cell nuclear extract for *in vitro* transcription human

Cat. No.	Amount
PR-777	200 $\mu$ l

For *in vitro* use only  
Quality guaranteed for 12 months  
Store at -80°C

### Avoid freeze / thaw cycles

### Form

Liquid. Supplied in 20 mM Tris-HCl, pH 8.0, 100 mM KCl, 0.2 mM EDTA, 1 mM DTT, 20% glycerol.

### Activity

1-5  $\mu$ l is sufficient for a gel mobility shift assay in a 20  $\mu$ l reaction, 5-10  $\mu$ l is sufficient for *in vitro* transcription assay and 20-50  $\mu$ l is sufficient for a protein-protein interaction assay.

### Application

The HeLa cell nuclear extract is specifically recommended for 1) *in vitro* transcription, 2) protein-DNA/RNA and protein-protein interactions, and 3) source of individual transcription factors.

### Purity

> 95% by SDS-PAGE.

### Description

The HeLa cell nuclear extract was prepared as described by Dignam et al. and Manley et al.. This extract contains all basal transcription factors, including TFIIA, -IIB, -IID, -IIE, -IIF, -IIH and RNA Polymerase II, as well as most gene-specific activators and cofactors, such as Sp1, Oct-1, NF- $\kappa$ B, USF, ATF, PC4, p300 etc. Therefore, it has been used as the source of transcription factors for the cell free transcription system to study specific transcription by RNA Polymerase II as well as RNA Polymerase III and for purifying transcription factors.

The HeLa cell nuclear extract is specifically recommended for 1) *in vitro* transcription; 2) protein- DNA/RNA and protein-protein interactions; and 3) source of individual transcription factors. Although this extract also contains most pre-mRNA processing factors, it is, however, not recommended for *in vitro* premRNA processing.

Please use our specific HeLa or 293 cell nuclear extract for the purpose of pre-mRNA processing.

### Selected References:

- Dignam *et al.* (1983) Accurate transcription initiation by RNA polymerase II in a soluble extract from isolated mammalian nuclei. *Nucleic Acids Res.* **11**:1475.
- Sawadogo *et al.* (1985) Factors involved in specific transcription by human RNA polymerase II: analysis by a rapid and quantitative *in vitro* assay. *Proc. Natl. Acad. Sci. USA* **82**:4394.
- Kadonaga *et al.* (1986) Affinity purification of sequence-specific DNA binding proteins. *Proc. Natl. Acad. Sci. USA* **83**:5889.
- Reinberg *et al.* (1987) Factors involved in specific transcription in mammalian RNA polymerase II. Functional analysis of initiation factors IIA and IID and identification of a new factor operating at sequences downstream of the initiation site. *J. Biol. Chem.* **262**:3322.
- Horikoshi *et al.* (1988) Transcription factor ATF interacts with the TATA factor to facilitate establishment of a preinitiation complex. *Cell* **54**:1033.
- Hoeffler *et al.* (1988) Activation of transcription factor IIIC by the adenovirus E1A protein. *Cell* **53**:907.
- Maldonado *et al.* (1996) Purification of human RNA polymerase II and general transcription factors. *Methods Enzymol.* **274**:72.