sequences. Addition of sigma factors will allow the enzyme to initiate RNA synthesis from specific bacterial and phage promoters. The core enzyme has a molecular weight of approximately 400 kDa.

**Source:** *E. coli* RNA Polymerase, Holoenzyme is isolated from *E. coli* strain BL21. The sigma factor 70 is purified from an *E. coli* strain carrying the cloned gene for sigma factor 70.

**Applications:**
- RNA synthesis from *E. coli* promoter
- Transcription initiation studies
- *In vitro* translation with PURExpress

**Reagents Supplied with Enzyme:**
5X *E. coli* RNA Polymerase Reaction Buffer

**Reaction Conditions:** 1X *E. coli* RNA Polymerase Reaction Buffer, supplemented with 0.5 mM each NTP and DNA template. Incubate at 37°C.

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**Quality Control Assays**

**DNA Endonuclease Activity:** Incubation of a 50 µl reaction containing 5 units of *E. coli* RNA Polymerase, Holoenzyme with 1 µg of X174 RF I DNA for 4 hours at 37°C resulted in less than 10% conversion to RF II, as determined by agarose gel electrophoresis.

**RNase Assay:** Incubation of a 10 µl reaction containing 1 unit of *E. coli* RNA Polymerase, Holoenzyme with 40 ng of RNA transcript for 4 hours at 37°C resulted in no detectable degradation, as determined by gel electrophoresis.

**DNA Exonuclease Activity:** Incubation of a 50 µl reaction containing 5 units of *E. coli* RNA Polymerase, Holoenzyme with 1 µg of a mixture of single- and double-stranded *H. coli* DNA for 4 hours at 37°C released < 0.1% of the total radioactivity.
Companion Products Sold Separately:

Ribonucleotide Solution Set
#N0450S 10 µmol of each
#N0450L 50 µmol of each

Ribonucleotide Solution Mix
#N0466S 10 µmol of each
#N0466L 50 µmol of each

RNase Inhibitor, Human Placenta
#M0307S 2,000 units
#M0307L 10,000 units

RNase Inhibitor, Murine
#M0314S 3,000 units
#M0314L 15,000 units

PURExpress® In Vitro Protein Synthesis Kit
#E6800S 10 reactions

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