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DATA QUALITY SHEET

T7 RNA Polymerase

T7 RNA Polymerase is a DNA-dependent RNA polymerase with strict specificity for its respective double-stranded promoters. It exhibits extremely high specificity for its cognate promoter sequences. Only T7 DNA or DNA cloned downstream from a T7 promoter can serve as a template for T7 RNA Polymerase-directed RNA synthesis.

It catalyzes the 5' \rightarrow 3' synthesis of RNA on either single-stranded DNA or double-stranded DNA downstream from its promoter.

Feature

• Incorporates modified nucleotides (e.g., aminoallyl-, biotin-, fluorescein-, digoxigenin-labeled nucleotides)

Applications

Synthesis of unlabeled and labeled RNA that can be used:

• For hybridization, in vitro RNA translation

• As aRNA, siRNA, substrate in RNase protection assays, template for genomic DNA sequencing

• In studies of RNA secondary structure and RNA-protein interactions, RNA splicing

Consensus promoter sequence:

T7: TAATACGACTCACTATAGGGAGA

QC Tests

Activity, SDS-PAGE/purity, DNase, RNase, endonuclease, transcription.

Source

Recombinant E. coli strain.

Storage Buffer

50mM Tris-HCl (pH 7.5 at 25°C), 1mM EDTA, 20mM 2-mercaptoethanol, 100mM NaCI, 0.1% (v/v) Triton® X-100 and 50% (v/v) glycerol.

Storage Conditions

Store at -20°C.

Unit Definition

One unit is defined as the amount of enzyme required to catalyze the incorporation of 1 nmol of NTP into acid-insoluble product in 60 minutes at 37° C in a total volume of 100µl. The reaction conditions are: 50mM Tris-HCl (pH 7.5 at 25°C), 6 mM MgCl₂,10 mM DTT, 2mM spermidine, 0.5mM each of ATP, GTP, CTP, and UTP, 0.5µCi [³H]CTP and 2µg DNA.