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New England Biolabs
Product Specification

Product Name:	*mRNA Decapping Enzyme*
Catalog #:	*M0608S*
Concentration:	*100,000 units/ml*
Unit Definition:	*One unit is defined as the amount of mRNA Decapping Enzyme required to convert 50% of a 500 nM m7G-capped substrate to a 5'-monophosphorylated form in a total reaction volume of 20 µl in 1 hour at 37°C.*
Shelf Life:	*24 months*
Storage Temp:	*-20°C*
Storage Conditions:	*10 mM Tris-HCl , 300 mM NaCl , 1 mM DTT , 0.1 mM EDTA , 50 % Glycerol, (pH 7.4 @ 25°C)*
Specification Version:	*PS-M0608S v2.0*
Effective Date:	*03 Aug 2020*

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 µl reaction in 1X mRNA Decapping Enzyme Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 units of mRNA Decapping Enzyme incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

Exonuclease Activity (Radioactivity Release) - A 50 µl reaction in 1X mRNA Decapping Enzyme Reaction Buffer containing 1 µg of a mixture of single and double-stranded [^{3}H] *E. coli* DNA and a minimum of 100 units of mRNA Decapping Enzyme incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

Phosphatase Activity (pNPP) - A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM $MgCl_2$ containing 2.5 mM *p*-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units of mRNA Decapping Enzyme incubated for 4 hours at 37°C yields <0.00001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.

Protein Purity Assay (SDS-PAGE) - mRNA Decapping Enzyme is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

RNase Activity (Extended Digestion) - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 100 units of mRNA Decapping Enzyme is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.

Date 03 Aug 2020

Derek Robinson
Director of Quality Control