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14 Nov 2013

Date

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## New England Biolabs Product Specification

Product Name: mRNA Cap 2 O Methyltransferase

Catalog #: M0366S

Concentration: 50,000 units/ml

Unit Definition: One unit is defined as the amount of enzyme required to methylate 10 pmoles of 80 nt long capped RNA transcript in 1 hour at

37°C.

Shelf Life: 24 months
Storage Temp: -20 °C

Storage Conditions: 100 mM NaCl, 20 mM Tris-HCl (pH 8.0), 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 0.1 % Triton®X-100

Specification Version: PS-M0366S v1.0
Effective Date: 14 Nov 2013

## Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50  $\mu$ l reaction in Capping Buffer containing 1  $\mu$ g of supercoiled PhiX174 DNA and a minimum of 50 units of mRNA Cap 2'-O-Methyltransferase 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  $\mu$ l reaction in Capping Buffer containing 1  $\mu$ g of a mixture of single and double-stranded [  $^3$ H] *E. coli* DNA and a minimum of 50 units of mRNA Cap 2'-O-Methyltransferase incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

**Protein Purity Assay (SDS-PAGE)** - mRNA Cap 2'-O-Methyltransferase is ≥ 99% 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 50 units of mRNA Cap 2'-O-Methyltransferase is incubated at 37°C. After incubation for 4 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.

Derek Robinson

Director of Quality Control

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