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## New England Biolabs Certificate of Analysis

Product Name: Vaccinia Capping System

Catalog Number: M2080S Concentration: 10,000 U/ml

Unit Definition: One unit of Vaccinia Capping Enzyme is defined as the amount of

enzyme required to incorporate 10 pmol of (α<sup>32</sup>P) GTP into an 80 nt

transcript in 1 hour at 37°C.

Packaging Lot Number: 10204928
Expiration Date: 09/2024
Storage Temperature: -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-M2080S v1.0

| Vaccinia Capping System Component List |                            |            |                      |  |
|--|----------------------------|------------|----------------------|--|
| <b>NEB Part Number</b>                 | Component Description      | Lot Number | Individual QC Result |  |
| N2080AVIAL                             | GTP                        | 10179511   | Pass                 |  |
| M2080SVIAL                             | Vaccinia Capping System    | 10182558   | Pass                 |  |
| B9003SVIAL                             | S-adenosylmethionine (SAM) | 10193027   | Pass                 |  |
| B2080AVIAL                             | 10X Capping Buffer         | 10164466   | Pass                 |  |

| Assay Name/Specification   | Lot # 10204928 |
|--|----------------|
| Endonuclease Activity (Nicking)  | Pass           |
| A 50 µl reaction in Capping Buffer containing 1 µg of supercoiled PhiX174 DNA and a  |                |
| minimum of 10 units of Vaccinia Capping System 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)   | Pass           |
| A 50 µl reaction in Capping Buffer containing 1 µg of a mixture of single and        |                |
| double-stranded [ 3H] E. coli DNA and a minimum of 10 units of Vaccinia Capping      |                |
| System incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.      |                |
| Protein Purity Assay (SDS-PAGE)  | Pass           |
| Vaccinia Capping System is ≥ 95% pure as determined by SDS-PAGE analysis using       |                |
| Coomassie Blue detection.  |                |
| RNase Activity (Extended Digestion)  | Pass           |
| A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA    |                |



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| Assay Name/Specification   | Lot # 10204928 |
|--|----------------|
| and a minimum of 10 units of Vaccinia Capping System 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.   |                |

This product has been tested and shown to be in compliance with all specifications.

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Jessica Cane Production Scientist 16 Nov 2022 Michael Tonello

Packaging Quality Control Inspector

27 Sep 2023

