

240 County Road Ipswich, MA 01938-2723

Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Certificate of Analysis

Product Name: Thermostable Inorganic Pyrophosphatase

Catalog Number: M0296S Concentration: 2,000 U/ml

Unit Definition: One unit is the amount of enzyme that will generate 1 μmol of

phosphate per minute from inorganic pyrophosphate under standard reaction conditions (a 10 minute reaction at 75°C in 50 mM Tricine [pH 8.5], 1 mM MgCl2, 0.32 mM PPi, reaction volume of 0.5 ml).

Packaging Lot Number: 10088332
Expiration Date: 10/2022
Storage Temperature: -20°C

Storage Conditions: 20 mM Tris-HCl, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol,

(pH 8.0 @ 25°C)

Specification Version: PS-M0296S/L v1.0

| Thermostable Inorganic Pyrophosphatase Component List |  |            |                      |  |
|---|--|------------|----------------------|--|
| <b>NEB Part Number</b>                                | Component Description                  | Lot Number | Individual QC Result |  |
| M0296SVIAL  | Thermostable Inorganic Pyrophosphatase | 10081158   | Pass                 |  |

| Assay Name/Specification   | Lot # 10088332 |
|--|----------------|
| dNTPase Activity A 500 μl reaction in CircumVent™ Sequencing Buffer in the presence of 200 μM each dNTPs and a minimum of 100 units Thermostable Inorganic Pyrophosphatase incubated for 1 hour at 75°C results in <0.01 μmole of inorganic phosphate from dNTPs as determined by the AAM assay.         | Pass           |
| Endonuclease Activity (Nicking) A 50 μl reaction in NEBuffer 2 containing 1 μg of supercoiled PhiX174 DNA and a minimum of 100 units of Thermostable Inorganic Pyrophosphatase incubated for 4 hours at 75°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass           |
| Exonuclease Activity (Radioactivity Release) A 50 μl reaction in NEBuffer 1 containing 1 μg of a mixture of single and double-stranded [ ³H] E. coli DNA and a minimum of 100 units of Thermostable Inorganic Pyrophosphatase incubated for 4 hours at 75°C releases <0.1% of the total radioactivity.   | Pass           |



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| Assay Name/Specification  | Lot # 10088332 |
|---|----------------|
| Phosphatase Activity (pNPP)   | Pass           |
| A 1 ml reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl2 containing 10 mM         |                |
| p-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units of Thermostable Inorganic |                |
| Pyrophosphatase incubated for 30 minutes at 75°C yields <0.00001 unit of alkaline   |                |
| phosphatase activity as determined by spectrophotometric analysis.                  |                |

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

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Timothy Meixsell Production Scientist 28 Nov 2020 Josh Hersey Packaging Quality Control Inspector

28 Nov 2020

