

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: α2-3,6,8 Neuraminidase

Catalog Number: P0720S
Concentration: 50,000 U/ml

Unit Definition: One unit is defined as the amount of enzyme required to cleave > 95%

of the terminal α-Neu5Ac from 1 nmol

Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-7-amino-4-methyl-coumarin

(AMC), in 5 minutes at 37°C in a total reaction volume of 10 μl.

Lot Number: 10018751
Expiration Date: 01/2020
Storage Temperature: -20°C

Storage Conditions: 50 mM NaCl, 20 mM Tris-HCl, 5 mM EDTA, (pH 7.5 @ 25°C)

Specification Version: PS-P0720S/L v1.0

| α2-3,6,8 Neuraminidase Component List |                        |            |                      |  |
|---------------------------------------|------------------------|------------|----------------------|--|
| <b>NEB Part Number</b>                | Component Description  | Lot Number | Individual QC Result |  |
| P0720SVIAL                            | α2-3,6,8 Neuraminidase | 0201801    | Pass                 |  |
| B1727SVIAL                            | 10X GlycoBuffer 1      | 10011906   | Pass                 |  |

| Assay Name/Specification  | Lot # 10018751 |
|---|----------------|
| Glycosidase Activity (β-Xylosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Xylosidase substrate (Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.              | Pass           |
| Glycosidase Activity (β1-3 Galactosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.  | Pass           |
| Glycosidase Activity (β1-4 Galactosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass           |



P0720S / Lot: 10018751

Page 1 of 4

| Assay Name/Specification   | Lot # 10018751 |
|--|----------------|
| Protease Activity (SDS-PAGE) A 20 μl reaction in 1X Glyco Buffer 1 containing 24 μg of a standard mixture of proteins and a minimum of 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C, results in no detectable degradation of the protein mixture as determined by SDS-PAGE with Coomassie Blue detection.                        | Pass           |
| Protein Purity Assay (SDS-PAGE) α2-3,6,8 Neuraminidase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.  | Pass           |
| Glycosidase Activity (β-N-Acetylglucosaminidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass           |
| Glycosidase Activity (α1-3 Galactosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-3Galβ1-4GlcNAc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                        | Pass           |
| Glycosidase Activity (α1-3 Mannosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Mannosidase substrate (Manα1-3Manβ1-4GlcNAc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                            | Pass           |
| Glycosidase Activity (α1-6 Galactosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-6Galα1-6Glcα1-2Fru-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                    | Pass           |
| Glycosidase Activity (α1-6 Mannosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Mannosidase substrate (Manα1-6Manα1-6(Manα1-3)Man-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                      | Pass           |
| Glycosidase Activity (β-Mannosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Mannosidase substrate (Manβ1-4Manβ1-4Man-AMC) and 500 units of α2-3,6,8   | Pass           |



P0720S / Lot: 10018751

Page 2 of 4

| Assay Name/Specification  | Lot # 10018751 |
|---|----------------|
| Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.  |                |
| Glycosidase Activity (β-N-Acetylgalactosaminidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.          | Pass           |
| Glycosidase Activity (Endo F1, F2, H) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F1, F2, H substrate (Dansylated invertase high mannose) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                           | Pass           |
| Glycosidase Activity (Endo F2, F3) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                                 | Pass           |
| Glycosidase Activity (PNGase F) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled PNGase F substrate (Fluoresceinated fetuin triantennary) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                                     | Pass           |
| Glycosidase Activity (α-Glucosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Glucosidase substrate (Glcα1-6Glcα1-4Glc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.   | Pass           |
| Glycosidase Activity (α-N-Acetylgalactosaminidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-N-Acetylgalactosaminidase substrate (GalNAcα1-3(Fucα1-2)Galβ1-4Glc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass           |
| Glycosidase Activity (α1-2 Fucosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-2Galβ1-4Glc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as   | Pass           |



P0720S / Lot: 10018751 Page 3 of 4

| Assay Name/Specification  | Lot # 10018751 |
|---|----------------|
| determined by thin layer chromatography.  |                |
| Glycosidase Activity (α1-3 Fucosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-3Galβ1-4GlcNAcβ1-3Galβ1-4Glc-AMC) and 500 units of α2-3,6,8 Neuraminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass           |

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

Alicia Bielik

**Production Scientist** 22 Jun 2018

Michael Tonello

Packaging Quality Control Inspector

30 Aug 2018



P0720S / Lot: 10018751

Page 4 of 4