

## New England Biolabs Certificate of Analysis

**Product Name:**  $\alpha$ 2-3,6,8,9 Neuraminidase A  
**Catalog Number:** P0722L  
**Concentration:** 20,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme required to cleave > 95% of the terminal  $\alpha$ -Neu5Ac from 1 nmol Neu5Ac $\alpha$ 2-3Gal $\beta$ 1-3GlcNAc $\beta$ 1-3Gal $\beta$ 1-4Glc-AMC, in 1 hour at 37°C in a total reaction volume of 10  $\mu$ l.  
**Lot Number:** 10020203  
**Expiration Date:** 08/2020  
**Storage Temperature:** -20°C  
**Storage Conditions:** 50 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, (pH 7.5 @ 25°C)  
**Specification Version:** PS-P0722S/L v1.0

| <b><math>\alpha</math>2-3,6,8,9 Neuraminidase A Component List</b> |                                    |                   |                             |
|--|------------------------------------|-------------------|-----------------------------|
| <b>NEB Part Number</b>   | <b>Component Description</b>       | <b>Lot Number</b> | <b>Individual QC Result</b> |
| P0722LVIAL   | $\alpha$ 2-3,6,8,9 Neuraminidase A | 10018781          | <b>Pass</b>                 |
| B1727SVIAL   | 10X GlycoBuffer 1                  | 10011906          | <b>Pass</b>                 |

| <b>Assay Name/Specification</b>   | <b>Lot # 10020203</b> |
|---|-----------------------|
| <p><b>Glycosidase Activity (Endo F1, F2, H)</b><br/>           A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F1, F2, H substrate (Dansylated invertase high mannose) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | <b>Pass</b>           |
| <p><b>Glycosidase Activity (Endo F2, F3)</b><br/>           A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>       | <b>Pass</b>           |
| <p><b>Glycosidase Activity (PNGase F)</b><br/>           A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled PNGase F substrate (Fluoresceinated fetuin triantennary) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>           | <b>Pass</b>           |

| Assay Name/Specification  | Lot # 10020203 |
|---|----------------|
| <p><b>Glycosidase Activity (<math>\alpha</math>-Glucosidase)</b><br/>A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-Glucosidase substrate (Glc<math>\alpha</math>1-6Glc<math>\alpha</math>1-4Glc-AMC) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>  | <b>Pass</b>    |
| <p><b>Glycosidase Activity (<math>\alpha</math>-N-Acetylgalactosaminidase)</b><br/>A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-N-Acetylgalactosaminidase substrate (GalNAc<math>\alpha</math>1-3(Fuca1-2)Gal<math>\beta</math>1-4Glc-AMC) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | <b>Pass</b>    |
| <p><b>Glycosidase Activity (<math>\alpha</math>1-2 Fucosidase)</b><br/>A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-Fucosidase substrate (Fuca1-2Gal<math>\beta</math>1-4Glc-AMC) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>  | <b>Pass</b>    |
| <p><b>Glycosidase Activity (<math>\alpha</math>1-3 Fucosidase)</b><br/>A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-Fucosidase substrate (Fuca1-3Gal<math>\beta</math>1-4GlcNAc<math>\beta</math>1-3Gal<math>\beta</math>1-4Glc-AMC) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>       | <b>Pass</b>    |
| <p><b>Glycosidase Activity (<math>\alpha</math>1-3 Galactosidase)</b><br/>A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-Galactosidase substrate (Gal<math>\alpha</math>1-3Gal<math>\beta</math>1-4GlcNAc-AMC) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>                               | <b>Pass</b>    |
| <p><b>Glycosidase Activity (<math>\alpha</math>1-3 Mannosidase)</b><br/>A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-Mannosidase substrate (Man<math>\alpha</math>1-3Man<math>\beta</math>1-4GlcNAc-AMC) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>                                   | <b>Pass</b>    |
| <p><b>Glycosidase Activity (<math>\alpha</math>1-6 Galactosidase)</b><br/>A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-Galactosidase substrate (Gal<math>\alpha</math>1-6Gal<math>\alpha</math>1-6Glc<math>\alpha</math>1-2Fru-AMC) and 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>        | <b>Pass</b>    |
| <p><b>Glycosidase Activity (<math>\alpha</math>1-6 Mannosidase)</b></p>   | <b>Pass</b>    |

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|--|----------------|
| <p>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 100 units of α2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>   |                |
| <p><b>Glycosidase Activity (β-Mannosidase)</b><br/>A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Mannosidase substrate (Manβ1-4Manβ1-4Man-AMC) and 100 units of α2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>                                  | <b>Pass</b>    |
| <p><b>Glycosidase Activity (β-N-Acetylgalactosaminidase)</b><br/>A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 100 units of α2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>   | <b>Pass</b>    |
| <p><b>Glycosidase Activity (β-N-Acetylglucosaminidase)</b><br/>A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 100 units of α2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | <b>Pass</b>    |
| <p><b>Glycosidase Activity (β-Xylosidase)</b><br/>A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Xylosidase substrate (Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC) and 100 units of α2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>                             | <b>Pass</b>    |
| <p><b>Glycosidase Activity (β1-3 Galactosidase)</b><br/>A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC) and 100 units of α2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>                 | <b>Pass</b>    |
| <p><b>Glycosidase Activity (β1-4 Galactosidase)</b><br/>A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 100 units of α2-3,6,8,9 Neuraminidase A incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>                | <b>Pass</b>    |
| <p><b>Protease Activity (SDS-PAGE)</b><br/>A 20 µl reaction in 1X Glyco Buffer 1 containing 24 µg of a standard mixture of</p>   | <b>Pass</b>    |

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|--|--------------------|
| <p>proteins and a minimum of 100 units of <math>\alpha</math>2-3,6,8,9 Neuraminidase A 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.</p> <p><b>Protein Purity Assay (SDS-PAGE)</b><br/> <math>\alpha</math>2-3,6,8,9 Neuraminidase A is <math>\geq</math> 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p> | <p><b>Pass</b></p> |

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