

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: α1-6 Mannosidase

Catalog Number: P0727S
Concentration: 40,000 U/ml

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

of the terminal α-D-mannose from 1 nmol of

Manα1-6Manα1-6Man-7-amino-4-methyl-coumarin (AMC), in 1 hour at 37°C

in a total reaction volume of 10 μl.

Lot Number:10053302Expiration Date:08/2020Storage Temperature:4°C

Storage Conditions: 50 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, (pH 7.5 @ 25°C)

Specification Version: PS-P0727S/L v1.0

| α1-6 Mannosidase Component List | | | | |
|---------------------------------|-----------------------|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| P0727SVIAL | α1-6 Mannosidase | 10053301 | Pass | |
| B9001SVIAL | Purified BSA | 10014762 | Pass | |
| B1727SVIAL | 10X GlycoBuffer 1 | 10041785 | Pass | |

| Assay Name/Specification | Lot # 10053302 |
|--|----------------|
| 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 80 units of α1-6 Mannosidase 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 80 units of α1-6 Mannosidase 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 80 units of α1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |



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| Assay Name/Specification | Lot # 10053302 |
| 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 80 units of α1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Glycosidase Activity (α-Neuraminidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Neuraminidase substrate (Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC) and 80 units of α1-6 Mannosidase 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 80 units of α1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| 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 80 units of α1-6 Mannosidase 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 80 units of α1-6 Mannosidase 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 80 units of α1-6 Mannosidase 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 80 units of α1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |



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| Assay Name/Specification | Lot # 10053302 |
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| 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 80 units of α 1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Glycosidase Activity (β-N-Acetylgalactosaminidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-N-Acetylgalactosaminidase substrate (GalNAc β 1-4Gal β 1-4Glc-AMC) and 80 units of α1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Glycosidase Activity (β-N-Acetylglucosaminidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAc β 1-4GlcNAc β 1-4GlcNAc-AMC) and 80 units of α1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Glycosidase Activity (β-Xylosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Xylosidase substrate (Xyl β 1-4Xyl β 1-4Xyl β 1-4Xyl-AMC) and 80 units of α1-6 Mannosidase 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 (Gal β 1-3GlcNAc β 1-4Gal β 1-4Glc-AMC) and 80 units of α1-6 Mannosidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F1, F2, H substrate (Dansylated invertase high mannose) and 80 units of α 1-6 Mannosidase 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 80 units of α 1-6 Mannosidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Protease Activity (SDS-PAGE) | Pass |



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| A 20 μl reaction in 1X Glyco Buffer 1 containing 24 μg of a standard mixture of proteins and a minimum of 400 units of α1-6 Mannosidase 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. | |
| Protein Purity Assay (SDS-PAGE) α1-6 Mannosidase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |

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

Tony Spear-Alfonso Production Scientist

12 Sep 2018

Michael Tonello

Packaging Quality Control Inspector

13 Sep 2019



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