

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:	α-N-Acetylgalactosaminidase
Catalog #:	P0734S/L
Concentration:	20,000 units/ml
Unit Definition:	One unit is defined as the amount of enzyme required to cleave > 95% of the terminal α -D-N-acetylgalactosamine from 1 nmol (GalNAc α 1-3)(Fuc α 1-2)Gal β 1-4Glc-7-amino-4-methyl-coumarin (AMC), in 1 hour at 37°C in a total reaction volume of 10 μ l.
Lot #:	0061603
Assay Date:	03/2016
Expiration Date:	3/2018
Storage Temp:	-20°C
Storage Conditions:	50 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, (pH 7.5 @ 25°C)
Specification Version:	PS-P0734S/L v1.0
Effective Date:	19 Feb 2016

Assay Name/Specification (minimum release criteria)	Lot #0061603
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 20 units of α - <i>N</i> -Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently- labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 20 units of α - <i>N</i> -Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled PNGase F substrate (Fluoresceinated fetuin triantennary) and 20 units of α - <i>N</i> -Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently- labeled β -Mannosidase substrate (Man β 1-4Man β 1-4Man-AMC) and 20 units of α -N-Acetylgalactosaminidase 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 20 units of α -N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass



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Glycosidase Activity (β-N-Acetylglucosaminidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β -N-Acetylglucosaminidase substrate (GlcNAc β 1-4GlcNAc β 1-4GlcNAc-AMC) and 20 units of α -N-Acetylgalactosaminidase 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 20 units of α -N-Acetylgalactosaminidase 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 20 units of α -N-Acetylgalactosaminidase 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 (Gal β 1-4GlcNAc β 1-3Gal β 1-4Glc -AMC) and 20 units of α -N-Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently- labeled α -Glucosidase substrate (Glc α 1-6Glc α 1-4Glc-AMC) and 20 units of α -N-Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently- labeled α -Neuraminidase substrate (Neu5Ac α 2-3Gal β 1-3GlcNAc β 1-3Gal β 1-4Glc-AMC) and 20 units of α -N- Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently- labeled α -Fucosidase substrate (Fuc α 1-2Gal β 1-4Glc-AMC) and 20 units of α -N-Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently- labeled α -Fucosidase substrate (Fuc α 1-3Gal β 1-4GlcNAc β 1-3Gal β 1-4Glc-AMC) and 20 units of α - <i>N</i> -Acetylgalactosaminidase 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 (minimum release criteria)	Lot #0061603 Pass
Glycosidase Activity (α1-3 Galactosidase) - A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Galactosidase substrate (Gal α 1-3Gal β 1-4GlcNAc-AMC) and 20 units of α -N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	
Glycosidase Activity (α1-3 Mannosidase) - A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Mannosidase substrate (Man α 1-3Man β 1-4GlcNAc-AMC) and 20 units of α -N-Acetylgalactosaminidase 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Galactosidase substrate (Gal α 1-6Gal α 1-6Glc α 1-2Fru-AMC) and 20 units of α -N-Acetylgalactosaminidase 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 (Man α 1-6Man α 1-6(Man α 1-3)Man-AMC) and 20 units of α -N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
Protease Activity (SDS-PAGE) - A 20 μ l reaction in 1X Glyco Buffer 1 containing 24 μ g of a standard mixture of proteins and a minimum of 100 units of α - <i>N</i> -Acetylgalactosaminidase 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) - α - <i>N</i> -Acetylgalactosaminidase is \geq 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass

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Authorized by Derek Robinson 19 Feb 2016



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Inspected by Jeremiah Read 18 Mar 2016