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## New England Biolabs Certificate of Analysis

Product Name:  $\beta$ -N-Acetylglucosaminidase S

Catalog Number: P0744L
Concentration: 4,000 U/ml

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

of the terminal, non-reducing β-N-Acetylglucosamine from 1 nmol

GlcNAcβ1-4GlcNAcβ1-4GlcNAc-7-amino-4-methylcoumarin (AMC), in 1 hour

at 37°C in a total reaction volume of 10 μl.

Packaging Lot Number: 10124273
Expiration Date: 05/2023
Storage Temperature: 4°C

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

Specification Version: PS-P0744S/L v1.0

β-N-Acetylglucosaminidase S Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
P0744LVIAL	β-N-Acetylglucosaminidase S	10153258	Pass	
B1727SVIAL	10X GlycoBuffer 1	10121467	Pass	

Assay Name/Specification	Lot # 10124273
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 16 units of β-N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S 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 # 10124273
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 16 units of β-N-Acetylglucosaminidase S 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 (Neu5Acα2-3Gal $\beta$ 1-3GlcNAc $\beta$ 1-3Gal $\beta$ 1-4Glc-AMC) and 16 units of $\beta$ -N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S 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 $β1-3$ GlcNAc $β1-4$ Gal $β1-4$ Glc-AMC) and 16 units of $β$ -N-Acetylglucosaminidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
<b>Glycosidase Activity (β-Mannosidase)</b> A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Mannosidase substrate (Man $\beta$ 1-4Man $\beta$ 1-4Man-AMC) and 16 units of β-N-Acetylglucosaminidase S 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 (GalNAcβ1-4Galβ1-4Glc-AMC) and 16 units of β-N-Acetylglucosaminidase S 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 (Galα1-3Gal $\beta$ 1-4GlcNAc-AMC) and 16 units of $\beta$ -N-Acetylglucosaminidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
Glycosidase Activity (α1-6 Galactosidase)	Pass



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Assay Name/Specification	Lot # 10124273
A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-6Galα1-6Glcα1-2Fru-AMC) and 16 units of β-N-Acetylglucosaminidase S 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 (Manα1-3Manβ1-4GlcNAc-AMC) and 16 units of β-N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S 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 16 units of β-N-Acetylglucosaminidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
Protein Purity Assay (SDS-PAGE) β-N-Acetylglucosaminidase S is ≥ 95% pure as determined by SDS-PAGE analysis using	Pass



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Coomassie Blue detection.	
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 24 units of β-N-Acetylglucosaminidase S 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

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

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Alicia Bielik **Production Scientist** 

24 May 2022

Erin Varney

Packaging Quality Control Inspector

24 May 2022



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