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## New England Biolabs Certificate of Analysis

Product Name: Endo H
Catalog Number: P0702S
Concentration: 500,000 U/ml

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

of the carbohydrate from 10 μg of denatured RNase B in 1 hour at 37°C in a total reaction volume of 10 μl (10 NEB units = 1 IUB

milliunit).

Packaging Lot Number: 10081123
Expiration Date: 08/2022
Storage Temperature: -20°C

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

Specification Version: PS-P0702S/L v1.0

Endo H Component List				
<b>NEB Part Number</b>	<b>Component Description</b>	Lot Number	Individual QC Result	
P0702SVIAL	Endo H	10081124	Pass	
B1720SVIAL	10X Glyco Buffer 3	10048943	Pass	
B1704SVIAL	Glycoprotein Denaturing Buffer	10065640	Pass	

Assay Name/Specification	Lot # 10081123
Glycosidase Activity (α1-6 Mannosidase) A 10 μl reaction in Glyco Buffer 3 containing 1 nM of fluorescently-labeled α-Mannosidase substrate (Μαηα1-6(Μαηα1-3)Μαη-ΑΜC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled β-Mannosidase substrate (Manβ1-4Manβ1-4Man-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled β-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 5,000 units of Endo H 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 # 10081123
Glycosidase Activity (β-N-Acetylglucosaminidase) A 10 μl reaction in Glyco Buffer 3 containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled β-Xylosidase substrate (Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 5,000 units of Endo H 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 µl reaction in 1X Glyco Buffer 3 containing 24 µg of a standard mixture of proteins and a minimum of 5,000 units of Endo H 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) Endo H is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
Glycosidase Activity (Endo F2, F3) A 10 μl reaction in Glyco Buffer 3 containing 1 nM of fluorescently-labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 5,000 units of Endo H incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
Glycosidase Activity (PNGase F)	Pass



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Assay Name/Specification	Lot # 10081123
A 10 μl reaction in Glyco Buffer 3 containing 1 nM of fluorescently-labeled PNGase F substrate (Fluoresceinated fetuin triantennary) and 5,000 units of Endo H incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	
<b>Glycosidase Activity (α-Glucosidase)</b> A 10 μl reaction in Glyco Buffer 3 containing 1 nM of fluorescently-labeled α-Glucosidase substrate (Glcα1-6Glcα1-4Glc-AMC) and 5,000 units of Endo H incubated for 20 hours at $37^{\circ}$ 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 3 containing 1 nM of fluorescently-labeled α-N-Acetylgalactosaminidase substrate (GalNAcα1-3(Fucα1-2)Galβ1-4Glc-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled α-Neuraminidase substrate (Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-2Galβ1-4Glc-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-3Galβ1-4GlcNAcβ1-3Galβ1-4Glc-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-3Galβ1-4GlcNAc-AMC) and 5,000 units of Endo H 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 3 containing 1 nM of fluorescently-labeled	Pass



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Assay Name/Specification	Lot # 10081123
α-Mannosidase substrate (Manα1-3Manβ1-4GlcNAc-AMC) and 5,000 units of Endo H incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	
Glycosidase Activity (α1-6 Galactosidase) A 10 μl reaction in Glyco Buffer 3 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-6Galα1-6Glcα1-2Fru-AMC) and 5,000 units of Endo H 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.

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Alicia Bielik Production Scientist 31 Aug 2020 Michael Tonello

Packaging Quality Control Inspector

31 Aug 2020



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