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New England Biolabs Certificate of Analysis

Product Name: Endo S
Catalog Number: P0741L
Concentration: 200,000 U/ml

He'l Deficition

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

of the carbohydrate from 5 µg of native mouse monoclonal IgG in 1

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

Packaging Lot Number: 10061225
Expiration Date: 12/2020
Storage Temperature: 4°C

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

Specification Version: PS-P0741S/L v2.0

Endo S Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
P0741LVIAL	Endo S	10061226	Pass	
B1727SVIAL	10X GlycoBuffer 1	10041785	Pass	

Assay Name/Specification	Lot # 10061225
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 2,000 units of Endo S 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 (Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC) and 2,000 units of Endo 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 (Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC) and 2,000 units of Endo 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)	Pass



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Assay Name/Specification	Lot # 10061225
A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 2,000 units of Endo S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	
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 2,000 units of Endo S 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 1 containing 24 µg of a standard mixture of proteins and a minimum of 2,000 units of Endo 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
Protein Purity Assay (SDS-PAGE) Endo S is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	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 2,000 units of Endo S 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 2,000 units of Endo 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) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-6Galα1-2Fru-AMC) and 2,000 units of Endo 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 2,000 units of Endo S incubated for 20 hours at 37°C results in no detectable activity as determined by	Pass



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Assay Name/Specification	Lot # 10061225
thin layer chromatography.	250 11 10001220
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 2,000 units of Endo S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
Functional Test (Magnetic Beads, Enzyme Removal) Magnetic chitin beads (50 µl) were equilibrated and incubated with 2,000 units of Endo S in 300 µl of 50mM ammonium formate, pH 4.4 . The beads were pelleted using a magnetic separation rack. No Endo S was detected in the supernatant as determined by activity assay and mass spectrometry analysis.	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 2,000 units of Endo 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 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 2,000 units of Endo 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 (Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC) and 2,000 units of Endo 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 2,000 units of Endo 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 2,000 units of Endo S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass



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

Alicia Bielik **Production Scientist**

26 Nov 2019

Michael Tonello

Packaging Quality Control Inspector 06 Dec 2019

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