

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: Remove-iT® PNGase F

Catalog Number: P0706L
Concentration: 225,000 U/ml

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

of the carbohydrate from 5 µg of DTT denatured RNase B in 1 hour at

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

Lot Number: 10042345
Expiration Date: 04/2020
Storage Temperature: 4°C

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

Specification Version: PS-P0706S/L v1.0

Remove-iT® PNGase F Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
P0706LVIAL	Remove-iT® PNGase F	10042346	Pass	
B3704SVIAL	10X GlycoBuffer 2	10040324	Pass	
B0706SVIAL	10X DTT	10039987	Pass	

Assay Name/Specification	Lot # 10042345
Glycosidase Activity (α-N-Acetylgalactosaminidase) A 10 μl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled α-N-Acetylgalactosaminidase substrate (GalNAcα1-3(Fucα1-2)Galβ1-4Glc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled α-Neuraminidase substrate (Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-2Galβ1-4Glc-AMC) and 450 units of Remove-iT® PNGase F 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 # 10042345
Glycosidase Activity (α1-3 Fucosidase) A 10 μl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-3Galβ1-4GlcNAcβ1-3Galβ1-4Glc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-3Galβ1-4GlcNAc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled α-Mannosidase substrate (Μαηα1-3Μαηβ1-4GlcNAc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-6Galα1-6Glcα1-2Fru-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled α-Glucosidase substrate (Glcα1-6Glcα1-4Glc-AMC) and 450 units of Remove-iT® PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
Endoglycosidase F1 Activity A 20 µl reaction in Glyco Buffer 2 containing 20 pmol of flourescently-labeled 2-AA Man-5 fluorescent standard and 1,125 units of Remove-iT® PNGase F incubated for 20 hours at 37°C results in no endoglycosidase F1 activity as determined by LC/MS analysis with fluorescent detection.	Pass
Functional Test (Magnetic Beads, Enzyme Removal) Magnetic chitin beads (50 µl) were equilibrated and incubated with 1,125 units of Remove-iT® PNGase F in 300 µl of 50 mM ammonium formate, pH 4.4 . The beads were pelleted using a magnetic separation rack. No Remove-iT® PNGase F was detected in the supernatant as determined by activity assay and mass spectrometry analysis.	Pass
Glycosidase Activity (Endo F1, F2, H)	Pass



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Assay Name/Specification	Lot # 10042345
A 10 µl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled Endo F1, F2, H substrate (Dansylated invertase high mannose) and 450 units of Remove-iT® PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	
Glycosidase Activity (Endo F2, F3) A 10 μl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled α-Mannosidase substrate (Manα1-6Manα1-6(Manα1-3)Man-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled 3-Mannosidase substrate (Manβ1-4Manβ1-4Man-AMC) and 450 units of Remove-iT® PNGase F ncubated 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 2 containing 1 nM of fluorescently-labeled 3-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled 3-Xylosidase substrate (Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC) and 450 units of Remove-iT® PNGase F 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 # 10042345
Glycosidase Activity (β1-3 Galactosidase) A 10 μl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 450 units of Remove-iT® PNGase F 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 2 containing 24 μg of a standard mixture of proteins and a minimum of 1,125 units of Remove-iT® PNGase F 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) Remove-iT® PNGase F 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.

Brad Landgraf
Production Scientist

14 Mar 2019

Michael Tonello

Packaging Quality Control Inspector

23 May 2019



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