

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/mI
Unit Definition:	One unit is defined as the amount of enzyme required to remove > 95% of the carbohydrate from 5 μ g of DTT denatured RNase B in 1 hour at 37°C in a total reaction volume of 10 μ l.
Lot Number:	10018032
Expiration Date:	07/2019
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	10015186	Pass	
B3704SVIAL	10X GlycoBuffer 2	0041709	Pass	
B0706SVIAL	10X DTT	0051802	Pass	

Assay Name/Specification	Lot # 10018032
Glycosidase Activity (β-N-Acetylgalactosaminidase) A 10 µl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled β -N-Acetylgalactosaminidase substrate (GalNAc β 1-4Gal β 1-4Glc-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 (GlcNAc β 1-4GlcNAc β 1-4GlcNAc-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 β-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 # 10018032
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
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 (α1-2 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 ncubated 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 (Manα1-3Manβ1-4GlcNAc-AMC) and 450 units of Remove-iT®	Pass





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PNGase F incubated for 20 hours at 37°C results in no detectable activity as	
determined by thin layer chromatography.	
Glycosidase Activity (α-Neuraminidase)	Pass
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.	
Glycosidase Activity (α-N-Acetylgalactosaminidase)	Pass
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.	
Glycosidase Activity (α-Glucosidase)	Pass
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.	
Glycosidase Activity (Endo F2, F3)	Pass
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.	
Glycosidase Activity (α1-6 Galactosidase)	Pass
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.	
Glycosidase Activity (α1-6 Mannosidase)	Pass
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 37ºC results in no detectable activity as	
determined by thin layer chromatography.	
Glycosidase Activity (β-Mannosidase)	Pass
A 10 μl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled β-Mannosidase substrate (Manβ1-4Manβ1-4Man-AMC) and 450 units of Remove-iT® PNGase	





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F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	
Glycosidase Activity (Endo F1, F2, H) 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.	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 ($50 \ \mu$ l) were equilibrated and incubated with 1,125 units of Remove-iT® PNGase F in 300 μ l 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

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

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Brad Landgraf Production Scientist 23 Jul 2018

Michae m. li

Michael Tonello Packaging Quality Control Inspector 15 Aug 2018

