NEW ENGLAND BioLabs Inc.
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New England Biolabs
Certificate of Analysis

Product Name:	*PNGase F (Glycerol-free), Recombinant*
Catalog Number:	*P0709L*
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. (65 NEB units = 1 IUB milliunit).*
Lot Number:	*10041608*
Expiration Date:	*04/2021*
Storage Temperature:	*4°C*
Storage Conditions:	*50 mM NaCl , 20 mM Tris-HCl , 5 mM EDTA, (pH 7.5 @ 25°C)*
Specification Version:	*PS-P0709S/L v1.0*

PNGase F (Glycerol-free), Recombinant Component List			
NEB Part Number	**Component Description**	**Lot Number**	**Individual QC Result**
P0709LVIAL	PNGase F (Glycerol-free), Recombinant	10041606	**Pass**
B3704SVIAL	10X GlycoBuffer 2	10040324	**Pass**
B2704SVIAL	NP-40	10021820	**Pass**
B1704SVIAL	Glycoprotein Denaturing Buffer	10043785	**Pass**

Assay Name/Specification	**Lot # 10041608**
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 5,000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC results in no detectable activity as determined by thin layer chromatography.	**Pass**
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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC results in no detectable	**Pass**

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Assay Name/Specification	Lot # 10041608
activity as determined by thin layer chromatography.	
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 5,000 of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 β-Mannosidase substrate (Manβ1-4Manβ1-4Man-AMC) and 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 β-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC results in no detectable activity as determined by thin layer chromatography.	**Pass**

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Assay Name/Specification	Lot # 10041608
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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC results in no detectable activity as determined by thin layer chromatography.	**Pass**
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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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 10,000 units of PNGase F (Glycerol-free), Recombinant 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) PNGase F (Glycerol-free), Recombinant 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 2 containing 1 nM of fluorescently-labeled α-N-Acetylgalactosaminidase substrate (GalNAcα1-3(Fucα1-2)Galβ1-4Glc-AMC) and 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC results in no detectable activity as determined by thin layer chromatography.	**Pass**
Glycosidase Activity (α-Neuraminidase)	**Pass**

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A 10 µl reaction in Glyco Buffer 2 containing 1 nM of fluorescently-labeled α-Neuraminidase substrate (Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC) and 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC results in no detectable activity as determined by thin layer chromatography.	
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 5000 units of PNGase F (Glycerol-free), Recombinant incubated for 20 hours at 37ºC 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.

Brad Landgraf
Production Scientist
09 Nov 2018

Jay Minichiello
Packaging Quality Control Inspector
06 Jun 2019