

New England Biolabs Certificate of Analysis

Product Name: PNGase F
Catalog Number: P0704S
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).
Packaging Lot Number: 10105452
Expiration Date: 01/2023
Storage Temperature: -20°C
Storage Conditions: 50 mM NaCl , 20 mM Tris-HCl , 5 mM EDTA , 50 % Glycerol, (pH 7.5 @ 25°C)
Specification Version: PS-P0704S/L v1.0

| PNGase F Component List | | | |
|-------------------------|--------------------------------|------------|----------------------|
| NEB Part Number | Component Description | Lot Number | Individual QC Result |
| P0704SVIAL | PNGase F | 10094768 | Pass |
| B3704SVIAL | 10X GlycoBuffer 2 | 10091370 | Pass |
| B2704SVIAL | NP-40 | 10098512 | Pass |
| B1704SVIAL | Glycoprotein Denaturing Buffer | 10090539 | Pass |

| Assay Name/Specification | Lot # 10105452 |
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| 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 5,000 units of 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 5,000 units of 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 5,000 units of | Pass |

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| <p>PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 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.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by</p> | Pass |

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| thin layer chromatography. | |
| <p>Protein Purity Assay (SDS-PAGE) PNGase F is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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(Fuca1-2)Galβ1-4Glc-AMC) and 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |

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| <p>Endoglycosidase F1 Activity (LC/MS) A 20 µl reaction in Glyco Buffer 2 containing 20 pmoles of 2-AA Man-5 fluorescent standard and 5,000 units of PNGase F incubated for 20 hours at 37°C results in no endoglycosidase F1 activity as determined by LC/MS analysis with fluorescent detection.</p> | Pass |
| <p>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 5,000 units of PNGase F incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |
| <p>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 incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> | Pass |

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

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Production Scientist
23 Apr 2021



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Packaging Quality Control Inspector
23 Apr 2021