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## New England Biolabs Certificate of Analysis

Product Name: β1-4 Galactosidase S

Catalog Number: P0745S
Concentration: 8,000 U/ml

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

of the terminal, β-D-galactose from 1 nmol Galβ1-4GlcNAcβ1-

3Galβ1-4Glc-7-amino-4-methyl-coumarin (AMC), in 1 hour at 37°C in a

total reaction volume of 10 μl.

Packaging Lot Number: 10220428
Expiration Date: 11/2025
Storage Temperature: -20°C

Storage Conditions: 50 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, (pH 7.5 @ 25°C)

Specification Version: PS-P0745S/L v1.0

| β1-4 Galactosidase S Component List |                       |            |                      |  |
|-------------------------------------|-----------------------|------------|----------------------|--|
| <b>NEB Part Number</b>              | Component Description | Lot Number | Individual QC Result |  |
| P0745SVIAL                          | β1-4 Galactosidase S  | 10217664   | Pass                 |  |
| B1727SVIAL                          | 10X GlycoBuffer 1     | 10181128   | Pass                 |  |

| Assay Name/Specification   | Lot # 10220428 |
|--|----------------|
| 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 80 units of β1-4 Galactosidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography. | Pass           |
| Glycosidase Activity (Endo F2, F3) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 80 units of β1-4 Galactosidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.       | Pass           |
| Glycosidase Activity (PNGase F) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled PNGase F substrate (Fluoresceinated fetuin triantennary) and 80 units of β1-4 Galactosidase S 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 # 10220428 |
|--|----------------|
| Glycosidase Activity ( $\alpha$ -Glucosidase) A 10 $\mu$ I reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled $\alpha$ -Glucosidase substrate (Glc $\alpha$ 1-6Glc $\alpha$ 1-4Glc-AMC) and 80 units of $\beta$ 1-4 Galactosidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                                  | 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 80 units of β1-4 Galactosidase 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 (Neu5Acα2-3Gal $\beta$ 1-3GlcNAc $\beta$ 1-3Gal $\beta$ 1-4Glc-AMC) and 80 units of $\beta$ 1-4 Galactosidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.                          | Pass           |
| <b>Glycosidase Activity (α1-2 Fucosidase)</b> A 10 $\mu$ I reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-2Gal $\beta$ 1-4Glc-AMC) and 80 units of $\beta$ 1-4 Galactosidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.   | Pass           |
| Glycosidase Activity ( $\alpha$ 1-3 Fucosidase) A 10 $\mu$ I reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled $\alpha$ -Fucosidase substrate (Fuc $\alpha$ 1-3Gal $\beta$ 1-4GlcNAc $\beta$ 1-3Gal $\beta$ 1-4Glc-AMC) and 80 units of $\beta$ 1-4 Galactosidase S 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 1 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-3Galβ1-4GlcNAc-AMC) and 80 units of β1-4 Galactosidase 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 80 units of β1-4 Galactosidase 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)  | Pass           |



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| Assay Name/Specification  | Lot # 10220428 |
|---|----------------|
| A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Galactosidase substrate (Galα1-6Galα1-6Glcα1-2Fru-AMC) and 80 units of β1-4 Galactosidase S incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.  |                |
| Glycosidase Activity (α1-6 Mannosidase) A 10 μl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Mannosidase substrate (Μαnα1-6Μαnα1-6(Μαnα1-3)Μαn-ΑΜC) and 80 units of β1-4 Galactosidase S 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 1 containing 1 nM of fluorescently-labeled β-Mannosidase substrate (Manβ1-4Manβ1-4Man-AMC) and 80 units of β1-4 Galactosidase S incubated 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 1 containing 1 nM of fluorescently-labeled β-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 80 units of β1-4 Galactosidase S 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 1 containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 80 units of β1-4 Galactosidase 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 80 units of β1-4 Galactosidase 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 80 units of β1-4 Galactosidase 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  | Pass           |



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| Frotein Funty Assay (3D3-FAGE)  |
|---|
| β1-4 Galactosidase S is ≥ 95% pure as determined by SDS-PAGE analysis using |
| Coomassie Blue detection.   |

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

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Alicia Bielik Production Scientist

22 Nov 2023

Michael Tonello

Packaging Quality Control Inspector

01 Dec 2023



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