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New England Biolabs Product Specification

Product Name: Boletopsis grisea Lectin (BGL)

Catalog #: P0867S

Concentration: 1 mg/ml

Shelf Life: 24 months

Storage Temp: -20°C

Storage Conditions: 50mM Tris-HCl, 200mM NaCl (pH 7.5 @, 25°C)

Specification Version: PS-P0867S v2.0

Effective Date: 01 Nov 2021

Assay Name/Specification (minimum release criteria)

Functional Testing (Epitope Directed Glycan Enrichment) - A 120 μ l reaction in 20 mM Tris-HCl pH 7.5 containing 100 ng of fluorescently labeled with procainamide G0/A2 N-glycan (asialo-, agalacto-, biantennary complex N-glycan) and 100 μ g BGL lectin incubated for 1.5 hours at 25°C results in \geq 85% enrichment as determined by UPLC-HILIC-FLR.

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 1 µl of Boletopsis grisea Lectin (BGL) 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 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (PNGase F) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled PNGase F substrate (Fluoresceinated fetuin triantennary) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (α -Glucosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Glucosidase substrate (Glc α 1-6Glc α 1-4Glc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (α -Neuraminidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Neuraminidase substrate (Neu5Ac α 2-3Gal β 1-3GlcNAc β 1-3Gal β 1-4Glc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.







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Glycosidase Activity ($\alpha 1$ -2 Fucosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Fucosidase substrate (Fuc $\alpha 1$ -2Gal $\beta 1$ -4Glc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity ($\alpha 1$ -3 Fucosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Fucosidase substrate (Fuc $\alpha 1$ -3Gal $\beta 1$ -4GlcNAc $\beta 1$ -3Gal $\beta 1$ -4Glc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity ($\alpha 1$ -3 Galactosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Galactosidase substrate (Gal $\alpha 1$ -3Gal $\beta 1$ -4GlcNAc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity ($\alpha 1$ -3 Mannosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Mannosidase substrate (Man $\alpha 1$ -3Man $\beta 1$ -4GlcNAc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (α 1-6 Galactosidase) - A 10 μ 1 reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Galactosidase substrate (Gal α 1-6Gal α 1-6Glc α 1-2Fru-AMC) and 1 μ 1 of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity ($\alpha 1$ -6 Mannosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -Mannosidase substrate (Man $\alpha 1$ -6Man $\alpha 1$ -6(Man $\alpha 1$ -3)Man-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (α -N-Acetylgalactosaminidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α -N-Acetylgalactosaminidase substrate (GalNAc α 1-3(Fuc α 1-2)Gal β 1-4Glc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (β -Mannosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β -Mannosidase substrate (Man β 1-4Man β 1-4Man-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (β -Xylosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β -Xylosidase substrate (Xyl β 1-4Xyl β 1-4Xyl β 1-4Xyl-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (β 1-3 Galactosidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β -Galactosidase substrate (Gal β 1-3GlcNAc β 1-4Gal β 1-4Glc-AMC) and 1 μ l of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.









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Glycosidase Activity (\(\beta 1-4 \) Galactosidase \) - A 10 \(\mu \) | reaction in Glyco Buffer 1 containing 1 \(\mu \) of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 1 μl of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

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 1 μl of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Glycosidase Activity (β -N-Acetylglucosaminidase) - A 10 μ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled β -N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 1 μl of Boletopsis grisea Lectin (BGL) incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.

Protease Activity (Non-Specific, SDS-PAGE) - A 20 µl reaction in 1X Glyco Buffer 2 containing 24 µg of a standard mixture of proteins and a minimum of 7 μl of Boletopsis grisea Lectin (BGL) was incubated for 20 hours at 37°C. After incubation, no detectable degradation of the protein mixture was determined by SDS-PAGE with Coomassie Blue detection.

Protein Purity Assay (SDS-PAGE) - Boletopsis grisea Lectin (BGL) is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

RNase Activity (Extended Digestion) - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl of Boletopsis grisea Lectin (BGL) is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.

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Derek Robinson

Director, Quality Control





