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New England Biolabs Certificate of Analysis

Product Name: Bacteroides Heparinase I

Catalog Number: P0735L
Concentration: 12,000 U/ml

Unit Definition: One unit is defined as the amount of enzyme that will liberate 1.0

µmol unsaturated oligosaccharides from porcine mucosal heparin per

minute at 30°C and pH 7.0 in a total reaction volume of 100 μl.

Packaging Lot Number: 10082465
Expiration Date: 07/2021
Storage Temperature: -80°C

Storage Conditions: 100 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, 5 mM CaCl2, (pH 7.5 @ 25°C)

Specification Version: PS-P0735S/L v1.0

| Bacteroides Heparinase I Component List | | | | |
|---|--|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| P0735LVIAL | Bacteroides Heparinase I | 10080839 | Pass | |
| B0735SVIAL | Bacteroides Heparinase Reaction Buffer (10X) | 10081039 | Pass | |

| Assay Name/Specification | Lot # 10082465 |
|---|----------------|
| Sulfatase Activity (2-0) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled 2-O-Sulfatase substrate (ΔUA2S-(1-4)-GlcNS6S-AMC) and 24 units of Bacteroides Heparinase I incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Protein Purity Assay (SDS-PAGE) Bacteroides Heparinase I is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |
| Protease Activity (SDS-PAGE) A 20 µl reaction in 1X Heparinase Reaction Buffer containing 24 µg of a standard mixture of proteins and a minimum of 120 units of Bacteroides Heparinase I 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 |
| Glycosidase Activity (β-N-Acetylglucosaminidase) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate | Pass |



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| Assay Name/Specification | Lot # 10082465 |
|---|----------------|
| (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 24 units of Bacteroides Heparinase I incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography. | |
| Glycosidase Activity (β-N-Acetylgalactosaminidase) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 24 units of Bacteroides Heparinase I incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Glycosidase Activity (β1-4 Galactosidase) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 24 units of Bacteroides Heparinase I incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Glycosidase Activity (β1-3 Galactosidase) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC) and 24 units of Bacteroides Heparinase I incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography. | Pass |
| Sulfatase and Uronidase Activity (N,6-O) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled N,6-O-Sulfatase substrate (ΔUA-(1-4)-GlcNS6S-AMC) and 24 units of Bacteroides Heparinase I incubated for 20 hours at 30°C 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.

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P0735L / Lot: 10082465

Alicia Bielik Production Scientist 18 Aug 2020

Michael Tonello

Packaging Quality Control Inspector

18 Aug 2020