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## New England Biolabs Certificate of Analysis

Product Name: Bacteroides Heparinase III

Catalog Number: P0737S
Concentration: 700 U/ml

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

µmol unsaturated oligosaccharides from heparan sulfate per minute at

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

Packaging Lot Number: 10204388
Expiration Date: 08/2024
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-P0737S/L v1.0

Bacteroides Heparinase III Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
P0737SVIAL	Bacteroides Heparinase III	10201509	Pass	
B0735SVIAL	Bacteroides Heparinase Reaction Buffer (10X)	10181104	Pass	

Assay Name/Specification	Lot # 10204388
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 1 unit of Bacteroides Heparinase III incubated for 20 hours at $30^{\circ}$ C results in no detectable activity as determined by thin layer chromatography.	Pass
Glycosidase Activity (β-N-Acetylglucosaminidase) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 1 unit of Bacteroides Heparinase III 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 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography.	Pass



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Assay Name/Specification	Lot # 10204388
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 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography.	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 5 units of Bacteroides Heparinase III 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) Bacteroides Heparinase III is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
Sulfatase Activity (2-O) A 10 μl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled 2-O-Sulfatase substrate (ΔUA2S-(1-4)-GlcNS6S-AMC) and 1 unit of Bacteroides Heparinase III 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 1 unit of Bacteroides Heparinase III 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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Maxwell/Elkus Production Scientist 04 Aug 2023 Michael Tonello

Packaging Quality Control Inspector

02 Nov 2023