

## 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  $\mu\text{mol}$  unsaturated oligosaccharides from porcine mucosal heparin per minute at 30°C and pH 7.0 in a total reaction volume of 100  $\mu\text{l}$ .  
**Packaging Lot Number:** 10081038  
**Expiration Date:** 07/2021  
**Storage Temperature:** -80°C  
**Storage Conditions:** 100 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, 5 mM CaCl<sub>2</sub>, (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)	10052584	Pass

Assay Name/Specification	Lot # 10081038
<p><b>Sulfatase Activity (2-O)</b>            A 10 <math>\mu\text{l}</math> reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled 2-O-Sulfatase substrate (<math>\Delta\text{UA}2\text{S}-(1-4)\text{-GlcNS6S-AMC}</math>) 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.</p>	Pass
<p><b>Sulfatase and Uronidase Activity (N,6-O)</b>            A 10 <math>\mu\text{l}</math> reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled N,6-O-Sulfatase substrate (<math>\Delta\text{UA}-(1-4)\text{-GlcNS6S-AMC}</math>) 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.</p>	Pass
<p><b>Glycosidase Activity (<math>\beta</math>1-3 Galactosidase)</b>            A 10 <math>\mu\text{l}</math> reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled <math>\beta</math>-Galactosidase substrate (<math>\text{Gal}\beta 1-3\text{GlcNAc}\beta 1-4\text{Gal}\beta 1-4\text{Glc-AMC}</math>) 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.</p>	Pass
<p><b>Glycosidase Activity (<math>\beta</math>1-4 Galactosidase)</b></p>	Pass

Assay Name/Specification	Lot # 10081038
<p>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.</p>	
<p><b>Glycosidase Activity (β-N-Acetylgalactosaminidase)</b> 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.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (β-N-Acetylglucosaminidase)</b> A 10 µl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (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.</p>	<b>Pass</b>
<p><b>Protease Activity (SDS-PAGE)</b> 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.</p>	<b>Pass</b>
<p><b>Protein Purity Assay (SDS-PAGE)</b> Bacteroides Heparinase I is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<b>Pass</b>

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

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30 Jul 2020



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