

## New England Biolabs Certificate of Analysis

**Product Name:** *Bacteroides Heparinase I*  
**Catalog Number:** P0735S  
**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:** 10180898  
**Expiration Date:** 02/2024  
**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
P0735SVIAL	Bacteroides Heparinase I	10180229	Pass
B0735SVIAL	Bacteroides Heparinase Reaction Buffer (10X)	10140899	Pass

Assay Name/Specification	Lot # 10180898
<p><b>Protease Activity (SDS-PAGE)</b>            A 20 <math>\mu\text{l}</math> reaction in 1X Heparinase Reaction Buffer containing 24 <math>\mu\text{g}</math> 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>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 (Gal<math>\beta</math>1-3GlcNAc<math>\beta</math>1-4Gal<math>\beta</math>1-4Glc-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 (<math>\beta</math>1-4 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 (Gal<math>\beta</math>1-4GlcNAc<math>\beta</math>1-3Gal<math>\beta</math>1-4Glc-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 (<math>\beta</math>-N-Acetylglucosaminidase)</b></p>	<b>Pass</b>

Assay Name/Specification	Lot # 10180898
<p>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>	
<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>Sulfatase and Uronidase Activity (N,6-O)</b> 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.</p>	<b>Pass</b>
<p><b>Sulfatase Activity (2-O)</b> 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.</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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Maxwell Elkus  
Production Scientist  
10 Feb 2023



Josh Hersey  
Packaging Quality Control Inspector  
13 Feb 2023