NEW ENGLAND BioLabs® Inc.

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New England Biolabs
Product Specification

Product Name:	*Bacteroides Heparinase I*
Catalog #:	*P0735S/L*
Concentration:	*12,000 units/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.*
Shelf Life:	*12 months*
Storage Temp:	*-80°C*
Storage Conditions:	*100 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, 5 mM $CaCl_2$, (pH 7.5 @ 25°C)*
Specification Version:	*PS-P0735S/L v1.0*
Effective Date:	*09 Dec 2015*

Assay Name/Specification (minimum release criteria)

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.

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.

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.

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 24 units of *Bacteroides* Heparinase I incubated for 20 hours at 30°C results in no detectable activity as determined by thin layer chromatography.

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.

Protein Purity Assay (SDS-PAGE) - *Bacteroides* Heparinase I is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

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Assay Name/Specification (minimum release criteria)
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 24 units of *Bacteroides* Heparinase I incubated for 20 hours at 30ºC results in no detectable activity as determined by thin layer chromatography.
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 30ºC results in no detectable activity as determined by thin layer chromatography.

Date 09 Dec 2015

Derek Robinson
Director of Quality Control

ISO 9001 Registered Quality Management
ISO 14001 Registered Environmental Management
ISO 13485 Registered Medical Devices