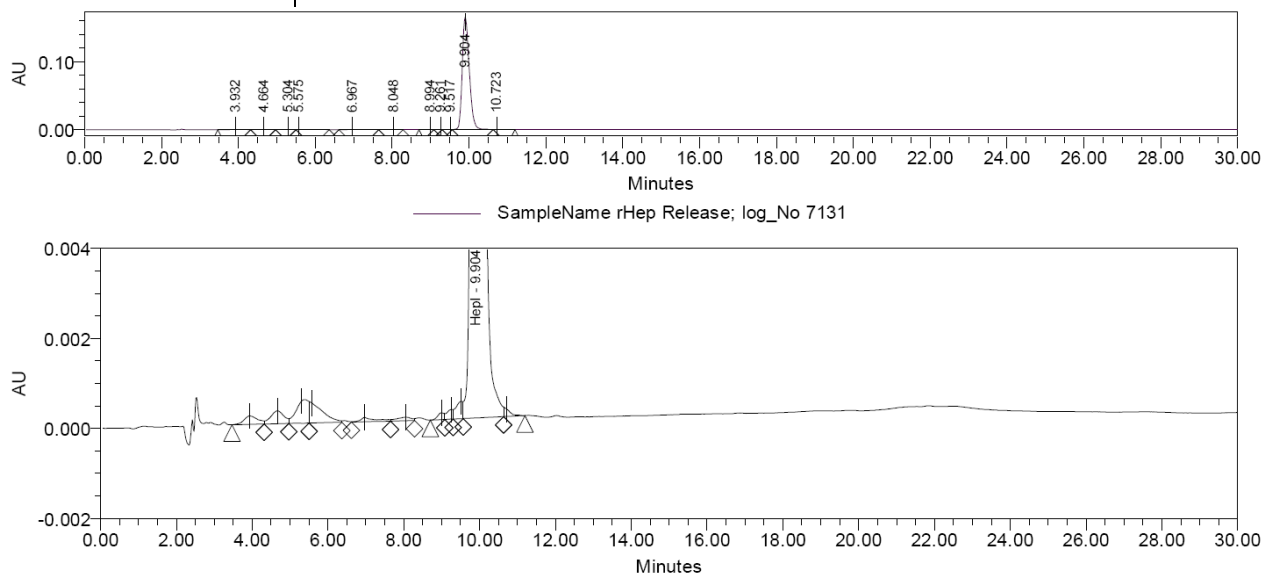
Data Sheet

Heparinase I | Diagnostic Grade

PN 50-008
50-009

Synonyms	Heparinase; heparin lyase; heparin eliminase
Source	*Flavobacterium heparinum* (recombinant)
EC Number	4.2.2.7
CAS Number	9025-39-2
Catalyzed Reaction	The enzyme cleaves selectively, via an elimination mechanism, highly sulfated polysaccharide chains containing 1-4 linkages between hexosamines and O-sulfated iduronic acid residues. The reaction yields oligosaccharide products (mainly disaccharides) containing unsaturated uronic acids which can be detected by UV spectroscopy at 232 nm. The enzyme also cleaves the antithrombin III binding pentasaccharide domain in the heparin molecule.
Substrate Specificity	Heparin; heparan sulfate (specific activity with heparin is approx. 3 times higher than with heparan sulfate).
Properties	• Molecular weight: 42,508 Da • Isoelectric point: 9.3 – 9.5 • pH optimum for activity: 6.5 – 7.5 • pH range for activity: 4 – 9 • Optimal testing temperature range: 20 °C – 37 °C • Optimal storage temperature: – 70 °C
Purity	≥95 % by reversed phase HPLC analysis.



IBEX Pharmaceuticals Inc.

5485 Paré, Suite 100, Montréal, Québec, Canada H4P 1P7
Tél : (514) 344-4004 Fax : (514) 344-8827 Website : www.IBEX.ca

Specific Activity

90-110 IU/mg using the following Unit definition.

One international unit (IU) is defined as the amount of enzyme that will liberate 1.0 μmole unsaturated oligosaccharides from porcine mucosal heparin per minute at 30 °C and pH 7.0.

(One Unit (U) is also defined in other preparation as 1 U forms 0.1 μmol of unsaturated uronic acid per hour; 1 IU is equivalent to 600 U).

Stability

Expiration is primarily established at 3 years after manufacturing. The Heparinase I is kept frozen at -70 ºC in aqueous buffers containing Sodium Phosphate or Sucrose 5%.

Applications

- For the neutralization of heparin in blood and plasma samples before analysis.
- For the similar in vitro neutralization of low molecular weight heparins.
- As integral part of in vitro diagnostic tests for the neutralization of heparin (blood clotting tests, platelet tests).
- For the preparation of low molecular weight heparins from unfractionated heparin.
- As a research reagent (glycosaminoglycan degradation).
- For the preparation of disaccharides of heparin and the preparation of oligosaccharide libraries.

Availability

A proprietary expression system for *F. heparinum* and the fermentation and isolation processes developed by IBEX Pharmaceuticals allow the production of large quantities of high purity product.

References

- Review: “Enzymatic Degradation of Glycosaminoglycans”. S. *Ernst et al. in Critical Reviews in Biochemistry and Molecular Biology (1995), 30(5): 387-444.*
- “Purification and Characterization of Heparin Lyases from *Flavobacterium heparinum*”. *D.L. Lohse and R.J. Linhardt in J. Biol. Chem. (1992) 267: 24347-24355.*
- “Purification and Characterization of Heparinase from *Flavobacterium heparinum*”. *V.C.Yang, R.J. Linhardt, H. Bernstein, C.L. Cooney and R. Langer in J. Biol. Chem. (1985) 260(3): 1849-1857.*
- “Substrate Specificity of the Heparin Lyases from *Flavobacterium heparinum*”. *U.R.Desai, H.Wang and R.J. Linhardt in Archives of Biochemistry and Biophysics (1993) 306(2): 461-468.*
- “Heparinase I from *Flavobacterium heparinum*. Mapping and Characterization of the Heparin Binding Domain”. *R. Sasisekharan, G. Venkataraman, R. Godavarti, S. Ernst, C.L. Cooney and R. Langer in J. Biol. Chem. (1996) 271 (6): 3124-3131.*

5485 Paré, Suite 100, Montréal, Québec, Canada H4P 1P7
Tél : (514) 344-4004 Fax : (514) 344-8827 Website : www.IBEX.ca

- "Cloning and Expression of Heparinase I Gene from *Flavobacterium heparinum*". *R. Sasisekharan, M. Bulmer, K.W. Moremen, C.L. Cooney and R. Langer in Proc. Natl. Acad. Sci. USA (1993) 90: 3660-3664.*

- "Neutralase (Heparinase I) as a Potential Heparin Reversal Agent in Coronary Artery Bypass Surgery". *P.J. Silver in Management of Bleeding in Cardiovascular Surgery, edited by R. Pifarré, MD, (2000) Hanley & Belfus, Inc., Philadelphia, PA.*

- "The effects of heparinase I and protamine on platelet reactivity". *T. Ammar and C.F. Fisher in Anesthesiology (1997) 86: 1382-1386.*

- "Heparinase I (Neutralase) Reversal of Systemic Anticoagulation". *L.G. Michelsen, M. Kikura, J.H. Levy et al. in Anesthesiology (1996) 85: 339-346.*

- "Neutralase Reverses the Anti-coagulant but not the Anti-thrombotic Activity of Heparin in a Rabbit Model of Venous Thrombosis". *P.J. Silver, R. Broughton, J. Bouthillier et al. in Thromb. Res. (1998) 91: 143-150.*

5485 Paré, Suite 100, Montréal, Québec, Canada H4P 1P7
Tél : (514) 344-4004 Fax : (514) 344-8827 Website : www.IBEX.ca