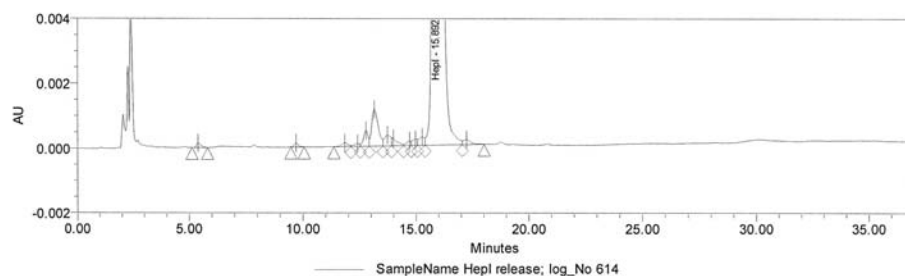
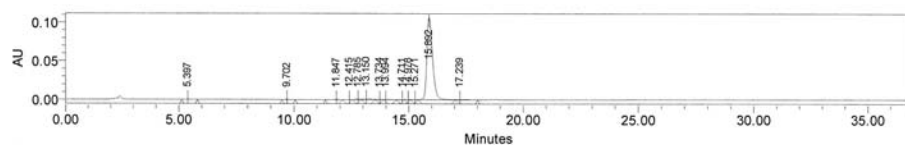


## Heparinase I

## Research Grade

PN 50-010  
50-010-001

<b>Synonyms</b>	Heparinase; heparin lyase; heparin eliminase
<b>Source</b>	<i>Flavobacterium heparinum</i> (recombinant)
<b>EC Number</b>	4.2.2.7
<b>CAS Number</b>	9025-39-2
<b>Catalyzed Reaction</b>	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.
<b>Substrate Specificity</b>	Heparin; heparan sulfate (specific activity with heparin is approx. 3 times higher than with heparan sulfate).
<b>Properties</b>	<ul style="list-style-type: none"> <li>• Molecular weight: 42,508 Da</li> <li>• Isoelectric point: 9.3 – 9.5</li> <li>• pH optimum for activity: 6.5 – 7.5</li> <li>• pH range for activity: 4 – 9</li> <li>• Optimal testing temperature range: 20 °C – 37 °C</li> <li>• Optimal storage temperature: – 70 °C</li> </ul>
<b>Purity</b>	≥95 % by reversed phase HPLC analysis.



IBEX Pharmaceuticals Inc.

## 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  $\mu$ mole unsaturated oligosaccharides from porcine mucosal heparin per minute at 30 °C and pH 7.0.

One Unit (U) is defined in other preparation as 1 U forms 0.1  $\mu$ mole of unsaturated uronic acid per hour at 25°C and pH 7.5; 1 IU is equivalent to 600 U

## Stability

- PN 50-010 (vial of 0.5 IU) – Expiration is 30 months from manufacturing date frozen at -70 °C in aqueous buffer containing Sodium Chloride, Sodium Phosphate and Sucrose 5%
- PN 50-010-001 (vial of 0.1 IU) – Expiration is 30 months from manufacturing date frozen at -70 °C in aqueous buffer containing Sodium Chloride, Sodium Phosphate and 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).
- In blood collection tubes for the neutralization of heparin.
- 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.

IBEX Pharmaceuticals Inc.

- “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.