

Product Information

INSULIN, HUMAN, RECOMBINANT EXPRESSED IN *E. coli*

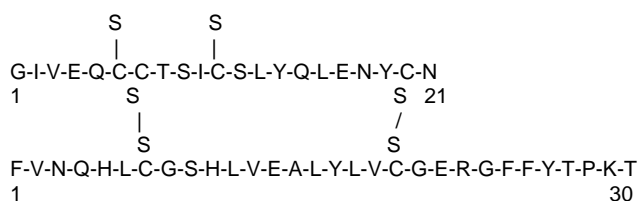
Product Number **I0259 and I2767**

Storage Temperature -0°C

CAS #: 11061-68-0

Synonyms: rDNA human insulin, recombinant human insulin, biosynthetic human insulin (BHI)

Structure¹



Product Description

Biosynthetic human insulin (BSI), or recombinant human insulin expressed in *E. coli* is of recombinant DNA origin. This recombinant human insulin is derived from proinsulin² and is chemically, physically and biologically identical to pancreatic human insulin.^{2,3,4,5}

Insulin is a 51 amino acid protein hormone produced *in vivo* in the pancreatic cells. The precursor protein (preproinsulin) contains a 23-30 amino acid signal peptide attached to the amino terminal of proinsulin. Proinsulin is composed of the insulin B-chain followed by a connecting peptide (C-peptide) and the A-chain. The signal peptide assists in translocating preproinsulin into the lumen of the endoplasmic reticulum, after which it is rapidly cleaved. Proinsulin is then transported to the Golgi complex where it is packaged into granules and converted to insulin. On secretion, equimolar amounts of insulin and C-peptide are released into the blood.^{6,7,8}

Insulin is the primary hormone responsible for controlling the cellular uptake, utilization and storage of glucose, amino acids, and fatty acids while inhibiting the breakdown of glycogen, protein, and fat. Several excellent reviews of the biochemistry, physiology, and pharmacology of insulin have been published.^{7,8,9,10}

Physical Properties

Appearance: White powder¹¹

Activity: Approx. 28 USP units/mg^{11,12}

Typical Zinc content: 0.3-0.6%¹³

Loss on Drying: Not more than 10%¹¹

Proinsulin content: <1 ppm¹³

Molecular Weight: ~6000¹

Isoelectric Point: 5.30-5.35¹

E^{1%} (276 nm): 10.4¹⁴ (9.6-11.2)¹³

Solubility: Clear colorless solution at 20 mg/mL in 0.01M HCl¹¹

Precautions and Disclaimer

Insulin, human, recombinant, expressed in *E. coli* is for laboratory use only, not for drug, household or other uses. Refer to the Material Safety Data Sheet (MSDS).

Preparation Instructions

Insulin has low solubility at neutral pH. It can be solubilized at 1-10 mg/mL in dilute acetic (1%) or hydrochloric acid, pH 2-3. Insulin can also be solubilized in 125 mM NaHCO₃.¹⁵ However, alkaline stock solutions are not recommended since high pH increases the rate of deamidation and aggregation.

Storage/Stability

Store the lyophilized powder below 0°C. A stock solution of insulin may be stored frozen at -20°C in single-use aliquots. Freeze-thaw cycles should be avoided. Alternatively, stock solutions can be stored for up to 6 months at 2-8°C if sterile filtered through a low protein binding membrane. Insulin solutions cannot be autoclaved. See Brange and Langkjoer, 1993 for a review on insulin structure and stability.¹⁶

Procedure

Recombinant human insulin or bovine insulin (such as Product Nos. I5500, I1882, I6634 and I4011) is often included as a medium supplement for cell culture. Insulin has long been recognized as a key factor in the regulation of the growth and differentiation of most cells *in vitro*. The concentration range is 1-10 µg/mL depending on the cell type.^{17,18} Methods for

immobilizing insulin on polystyrene dishes have been proposed for applications involving protein-free cell

Sigma also offers a murine monoclonal antibody to human insulin, Product No. I2018.

References

1. Merck Index 12, 5011 (1996).
2. Chance, RE and Frank, BH. Research, Development, Production, and Safety of Biosynthetic Human Insulin, *Diabetes Care* 16 (Suppl 3), 133-142 (1993).
3. Johnson, IS. Authenticity and purity of human insulin (recombinant DNA). *Diabetes Care* 5, (Suppl. 2), 4-12 (1982).
4. Chance, RE, et al. Chemical, physical, and biological properties of recombinant human insulin. In *Insulins, Growth Hormone, and Recombinant DNA Technology*. Gueriguian, JL, Ed. New York, Raven, p. 71-86 (1981).
5. Chance, RE, et al. Chemical, physical, and biologic properties of biosynthetic human insulin. *Diabetes Care* 4, 147-154 (1981).
6. Nolan, C. et al., *J. Biol. Chem.*, 246, 2780-2795 (1971).
7. Davis, S.N., and Granner, D.K., (1996) In J.G. Hardman, et al. (eds), *Goodman & Gilman's The Pharmacological Basis of Therapeutics*. Ninth Edition. New York: McGraw-Hill. pp. 1487-1517.
8. Wallis, M. et al. (1985) *The Biochemistry of the Polypeptide Hormones*. New York: John Wiley & Sons. pp 256-300.
9. Czech, M.P. (Ed.) *Molecular Basis of Insulin Action*. New York, Plenum Press (1985).
10. Kono, T., *Vitamins & Hormones*, 44, 103-154 (1988).
11. Sigma product specifications
12. *US Pharmacopoeia* (USP) 23 (1995) p 807.
13. Supplier Information
14. Fasman, GD. *CRC Practical Handbook of Biochemistry and Molecular Biology*, CRC Press, Boston, p. 270, 1989. (Product No. C3671)
15. Lougheed, W.D. et al., *Diabetologia*, 20, 51-53 (1981).
16. Brange J, et al. Insulin Structure and Stability. *Pharm Biotechnol.* 5, 315-350 (1993).
17. Freshney, R.I. (1994) *Culture of Animal Cells*, Third Edition. New York:Wiley-Liss.
18. Minamoto, Y. et al., *Cytotechnol.*, 5 Suppl. 2, S35-S51 (1991).
19. Ito, Y. et al. Photoimmobilization of insulin on polystyrene dishes for protein-free cell culture. *Biotechnol. Prog.*, 12(5), 700-702 (1996).

kat 1/21/99

Sigma brand products are sold through Sigma-Aldrich, Inc.

Sigma-Aldrich, Inc. warrants that its products conform to the information contained in this and other Sigma-Aldrich publications. Purchaser must determine the suitability of the product(s) for their particular use. Additional terms and conditions may apply. Please see reverse side of the invoice or packing slip.