

## Product Information

### Hirudin recombinant

Catalog Number **H0393**  
Storage Temperature  $-20\text{ }^{\circ}\text{C}$

CAS RN 8001-27-2 (native form)

#### Product Description

The anticoagulant hirudin is the most potent natural inhibitor of both soluble and clot-bound thrombin. Hirudin occurs naturally in leeches (*Hirudo medicinalis*).<sup>1,2</sup> Hirudin binds thrombin at 1:1 stoichiometry with high affinity, and covers more than 20% of the surface area of thrombin, occluding both the active site and exosite I (fibrinogen and PAR recognition site). This coverage blocks thrombus growth and platelet activation.<sup>1</sup> Hirudin is not metabolized in the bloodstream of humans and is eliminated unchanged via kidney filtration.

Hirudin is a ~7 kDa acidic protein containing 65 amino acid residues. Native hirudin contains a sulfated tyrosyl residue (Tyr<sup>63</sup>), three disulfide bridges, and a high proportion of dicarboxylic acids.<sup>3-5</sup> Hirudin is not glycosylated and lacks tryptophan, arginine, and methionine residues. At least 20 isoforms have been identified and sequenced.<sup>5</sup>

Hirudin variant 1 (HV-1) is a recombinant protein, produced from cDNA expressed in a proprietary host. This product corresponds to the HV-1 variant sequence, except the Tyr<sup>63</sup> residue is not sulfated.<sup>1</sup> This product is supplied as a powder, containing glycine as a bulk stabilizer.

Specific Activity:  $\geq 7,000$  antithrombin units/mg protein

One antithrombin unit (ATU) will neutralize one NIH unit of thrombin at  $37\text{ }^{\circ}\text{C}$ , based on direct comparison to an NIH thrombin reference standard.

#### Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

#### Preparation Instructions

Hirudin is soluble in water. The literature cites the use of "Dilution Fluid II" (35.7 mM acetic acid, 35.7 mM sodium diethyl barbiturate, 0.85% NaCl, 1% bovine serum albumin, and 0.5% PEG, pH 7.4) to dissolve hirudin (500 ATU/mL) and thrombin.<sup>6</sup> Hirudin is reported to be soluble in pyridine, but practically insoluble in alcohol, ether, acetone, or benzene.<sup>7</sup>

Stability testing of frozen solutions of this product has not been performed in our laboratories. One published reference cites storage of 1.0 mg/mL stock solutions of recombinant hirudin in saline at  $-30\text{ }^{\circ}\text{C}$ .<sup>8</sup>

#### Storage/Stability

Store the product at  $-20\text{ }^{\circ}\text{C}$ .

#### References

1. Markwardt, F., *Semin. Thromb. Hemost.*, **28(5)**, 405-414 (2002).
2. Walsmann, P., *Semin. Thromb Hemost.*, **17(2)**, 83-87 (1991).
3. Markwardt, F., and Walsmann, P., *Hoppe-Seyler's Z. Physiol. Chem.*, **312(1-3)**, 85-98 (1958).
4. Dodt, J. *et al.*, *FEBS Lett.*, **165(2)**, 180-184 (1984).
5. Stone, S.R., and Maraganore, J.M., *Methods Enzymol.*, **223**, 312-336 (1993).
6. Loison, G. *et al.*, *Nat. Biotech.*, **6**, 72-77 (1988).
7. The Merck Index, 11th ed., Entry# 4638 (1989).
8. Jiang, S.-Y. *et al.*, *PLoS One*, **8(6)**, e64336 (2013).

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