

RayBiotech, Inc.

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Certificate of Analysis and Data Sheet

Recombinant Ovine Leptin (Anti-Obesity Protein) {rOLeptin (AOBP)}

Source: E.Coli	Catalog No. IP-01-239
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Description :

Recombinant Ovine Leptin produced in E.Coli is a single, non-glycosylated, polypeptide chain containing 146 amino acids and having a molecular mass of 16,130 Dalton.

The rOLeptin is purified by proprietary chromatographic techniques.

Physical Appearance:

Sterile Filtered White lyophilized (freeze-dried) powder.

Formulation:

The protein was lyophilized from a concentrated (1mg/ml) solution with 0.0045mM NaHCO₃.

Solubility:

It is recommended to reconstitute the lyophilized rOLeptin in sterile 18M-cm H₂O not less than 100µg/ml, which can then be further diluted to other aqueous solutions.

Stability:

Lyophilized rOLeptin although stable at room temperature for 3 weeks, should be stored desiccated below -18°C Upon reconstitution rOLeptin should be stored at 4°C between 2-7 days and for future use below -18°C. For long term storage it is recommended to add a carrier protein (0.1% HSA or BSA).

Please prevent freeze-thaw cycles.

Purity:

Greater than 95.0% as determined by:

- (a) Analysis by RP-HPLC.
- (b) Anion-exchange FPLC.
- (c) Analysis by reducing and non-reducing SDS-PAGE Silver Stained gel.

Amino acid sequence:

The sequence of the first five N-terminal amino acids was determined and was found to be Ala-Val-Pro-Ile-Arg.

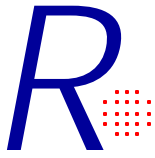
Dimers and aggregates:

Less than 1% as determined by silver-stained SDS-PAGE gel analysis.

Biological Activity:

rOLeptin is fully biologically active when compared to standards. The ED₅₀, calculated by the leptin-dependant stimulation of Human OB-R transfected murine BaF3 indicator cells is 0.33-0.05 ng/ml.

Endotoxin:



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Less than 0.1 ng/ μ g (IEU/ μ g) of rOLEptin.

Protein content:

Protein quantitation was carried out by two independent methods:

1. UV spectroscopy at 280 nm using the absorbency value of 0.2 as the extinction coefficient for a 0.1% (1mg/ml) solution. This value is calculated by the PC GENE computer analysis program of protein sequences (IntelliGenetics).
2. Analysis by RP-HPLC, using calibrated solution of Ovine Leptin as a Reference Standard.

Usage:

This material is offered for research, laboratory or further evaluation purposes.