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Not for use in diagnostic procedures.



Interleukin-6, human (hIL-6) recombinant (*E. coli*)

 **Version: 09**

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Cat. No. 11 138 600 001 200,000 U
2 µg, 1 ml

Store product at –15 to –25°C.

1.	General Information	3
1.1.	Contents	3
1.2.	Storage and Stability	3
	Storage Conditions (Product)	3
1.3.	Additional Equipment and Reagent required	3
1.4.	Application	3
2.	How to Use this Product	4
2.1.	Before you Begin	4
	General Considerations	4
	Primary structure	4
	Working Solution	4
2.2.	Protocols	4
	Cultivation of mouse-mouse-hybridoma cells	4
2.3.	Parameters	5
	Molecular Weight	5
	Purity	5
	Specific Activity	5
	Specificity	5
	Unit Definition	5
	EC ₅₀ definition	5
	Working Concentration	5
3.	Results	6
	Comparison of proliferation response of 7TD1 cells	6
4.	Additional Information on this Product	7
4.1.	Test Principle	7
	Preparation	7
4.2.	Quality Control	7
5.	Supplementary Information	8
5.1.	Conventions	8
5.2.	Changes to previous version	8
5.3.	Trademarks	9
5.4.	License Disclaimer	9
5.5.	Regulatory Disclaimer	9
5.6.	Safety Data Sheet	9
5.7.	Contact and Support	9

1. General Information

1.1. Contents

Vial / Bottle	Cap	Label	Function / Description	Content
1	red	Interleukin-6, human (hIL-6)	<ul style="list-style-type: none"> Solution, filtered through 0.2 µm pore size membrane. 200,000 U/ml (2 µg/ml) in PBS (phosphate buffered saline) and 1 mg/ml BSA (bovine serum albumin). <p>i Purity of BSA: >98%, endotoxin (LAL): <1 EU/mg BSA.</p>	1 bottle, 1 ml

1.2. Storage and Stability

Storage Conditions (Product)

The product is shipped on dry ice.

When stored at –15 to –25°C, the product is stable through the expiration date printed on the label.

Vial / Bottle	Cap	Label	Storage
1	red	Interleukin-6, human (hIL-6)	Store in aliquots at –15 to –25°C. ⚠️ Avoid repeated freezing and thawing.

1.3. Additional Equipment and Reagent required

For cultivation of mouse-mouse-hybridoma cells

- HAT selection medium, such as RPMI 1640, containing 10% FCS (v/v), 2 mM L-glutamine, 1x non-essential amino acids, 1 mM sodium pyruvate, 50 µM 2-mercaptoethanol, and 1x HAT-media supplement.
- HT medium, such as RPMI 1640, containing 10% FCS (v/v), 2 mM L-glutamine, 1x non-essential amino acids, 1 mM sodium pyruvate, 50 µM 2-mercaptoethanol, 1x HT-media supplement, and 100 U/ml hIL-6.

1.4. Application

Recombinant IL-6, human is a growth factor for murine and human B cells. It can be used to replace feeder cells in the preparation of murine and human hybridomas.

2. How to Use this Product

2.1. Before you Begin

General Considerations

Primary structure

One polypeptide chain (184 amino acids) is identical to that of natural human IL-6, but contains an altered sequence within the first 15 amino-terminal amino acids.

Working Solution

Dilute the concentrated Interleukin-6 solution (200,000 U/ml) with PBS or culture medium containing 1 mg/ml BSA or HSA (human serum albumin) or 1 to 10% serum.

2.2. Protocols

Cultivation of mouse-mouse-hybridoma cells

- 1 For selection of freshly fused mouse-mouse hybridoma cells, add 100 U/ml hIL-6 to HAT selection medium, see section, **Additional Equipment and Reagent Required**.

- 2 Cultivate cells for 5 to 7 days.

- 3 Feed cells by carefully aspirating 100 μ l from the supernatant and adding 100 μ l of HAT selection medium containing hIL-6.

- 4 Cultivate cells for 3 days.

- 5 Feed cells as described above with HT medium, see section, **Additional Equipment and Reagent Required**.

- 6 Cultivate cells for 7 to 14 days and feed cells with the HT medium as required.

- 7 For cloning of freshly produced hybridoma cells or for subcultivation or recloning of established mouse-mouse hybridoma lines, add 100 U/ml hIL-6 to the culture medium, for example, RPMI 1640, 10% FCS (v/v), 2 mM L-glutamine, 1x non-essential amino acids, 1 mM sodium pyruvate, and 50 μ M 2-mercaptoethanol.

2.3. Parameters

Molecular Weight

20,600 Da

Purity

≥95% pure as determined by HPLC or SDS-PAGE.

Endotoxin level: ≤10 EU/ml (LAL).

Specific Activity

≤100 MU/mg

(hIL-6, NIBSC standard 88/514), at least the same specific activity (EC_{50}) compared to the indicated standard is guaranteed, see section, **Results, Figure 1**.

Specificity

Recombinant IL-6, human is effective on mouse and human cells.

Unit Definition

EC_{50} definition

The amount of hIL-6 that is required to support half-maximal stimulation of cell proliferation (MTT cleavage) with 7TD1 cells (1 unit equals <0.01 ng).

Working Concentration

Interleukin-6 exerts its biological activity in the concentration range of 10 to 100 U/ml (0.1 to 1 ng/ml). Use a concentration of 100 U/ml (1 ng/ml) for the preparation of B-cell hybridomas.

3. Results

Comparison of proliferation response of 7TD1 cells

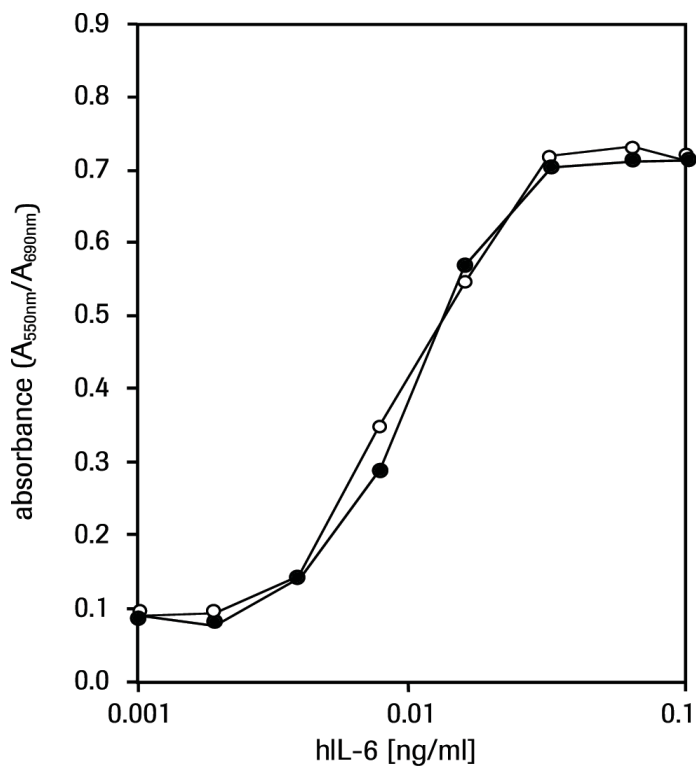


Fig. 1: Proliferation of 7TD1 cells (mouse-mouse hybridoma) in response to recombinant human interleukin-6 (hIL-6) (▪), or the hIL-6 reference standard (NIBSC standard 88/514) (○).

4. Additional Information on this Product

4.1. Test Principle

IL-6, a 20.6 KDa protein, also known as B-cell stimulatory factor 2 (BSF-2, BCDF), interferon- β 2 (IFN- β 2), hybridoma/plasmacytoma growth factor (H(P)GF), and hepatocyte-stimulating factor is produced by many cell types, such as T cells, monocytes, fibroblasts, and endothelial cells. IL-6 induces the terminal maturation of activated B cells into antibody-producing cells (B-cell differentiation activity). The hepatic acute phase response is induced by IL-6 (hepatocyte-stimulating activity). It was shown that IL-6 and IL-3 act synergistically to support the proliferation of hematopoietic stem cells (hemapoietic activity), however, IL-6 does not have antiviral activity, although it has been called IFN-2. High affinity receptors for IL-6 are found on many cell types.

Preparation

Interleukin-6 is produced in *E. coli* and purified by standard chromatographic techniques.

4.2. Quality Control

For lot-specific certificates of analysis, see section **Contact and Support**.

5. Supplementary Information

5.1. Conventions

To make information consistent and easier to read, the following text conventions and symbols are used in this document to highlight important information:

Text convention and symbols

i *Information Note: Additional information about the current topic or procedure.*

⚠ **Important Note: Information critical to the success of the current procedure or use of the product.**

① ② ③ etc. Stages in a process that usually occur in the order listed.

① ② ③ etc. Steps in a procedure that must be performed in the order listed.

* (Asterisk) The Asterisk denotes a product available from Roche Diagnostics.

5.2. Changes to previous version

Layout changes.
Editorial changes.

5.3. Trademarks

All product names and trademarks are the property of their respective owners.

5.4. License Disclaimer

For patent license limitations for individual products please refer to:

List of biochemical reagent products.

5.5. Regulatory Disclaimer

For life science research only. Not for use in diagnostic procedures.

5.6. Safety Data Sheet

Please follow the instructions in the Safety Data Sheet (SDS).

5.7. Contact and Support

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support Site.**

To call, write, fax, or email us, visit **sigma-aldrich.com**, and select your home country. Country-specific contact information will be displayed.

