PeliKine compact™ human IL-13

Sanquin

Specification sheet

REF M1913

Introduction

Interleukin 13 (IL-13) is an immunoregulatory protein produced by activated T-cells. The protein encoded by the IL-13 cDNA is the human homologue of a mouse TH2 product called P600 [1-3]. IL-13 shares many of its biological activities with the TH2 cytokine Interleukin 4; both cytokines are able to enhance expression of CD23 on monocytes and B-cells and also induce IgE production [3-8]. Production of many LPS-induced monokines, such as IL-1α, IL-1β, IL-6, IL-8, IL-10, TNFα, IFNα, MIP-1α, GM-CSF and G-CSF are inhibited by IL-13 [3,6], whereas IL-1ra is upregulated. These properties are shared with IL-4 and IL-10. Therefore IL-4 and IL-13 can be considered as anti-inflammatory molecules. In contrast to IL-4, IL-13 has no growth-promoting effect on T-cells and cannot compete for IL-4 binding to a human T-cell line. Therefore it was thought that the specific receptor for IL-13 is lacking on T-cells [4,9]. However, recently an inhibitory effect of IL-13 on IL-8- and RANTES-induced chemotaxis of T-cells has been described, indicating that T-cells do respond to IL-13 [10], possibly by inhibition of production of the TH1 inducer IL-12 [4].
Bioassays for the quantification of IL-13, including the proliferative assay of an IL-13- dependent subclone of the B9 cell line [12] can be used. However, IL-13 shares many of the biological effects of IL-4 and for this reason the two commonly interfere in bioassays.
Furthermore, these bioassay, although sensitive, are time consuming and susceptible to interference by other substances.
The PeliKine compact™ human IL-13 ELISA kit [11] has been developed for faster, more reproducible and specific quantification of human IL-13 in serum, plasma and other body fluids, as well as in cell-culture supernatant.

Assay procedure

See Assay procedure for PeliKine™ compact ELISA kit.

Kitcomponent list

Quantity	Kit component		Volume	Cap colour
1 vial	coating antibody	100-fold concentrated	375 µl	red
1 vial	blocking reagent	50-fold concentrated	2 ml	transparent
1 vials	IL-13 standard (lyophilized)	see label	500 µl	black
1 vial	biotinylated antibody	100-fold concentrated	375 µl	yellow
1 vial	streptavidin-poly-HRP conjugate	10,000-fold concentrated	20 µl	brown
1 bottle	HPE-dilution buffer	5-fold concentrated	55 ml	
3 pcs	microtiter plate + lid	-	-	
10 pcs	plate seals	-	-	

Sensitivity

MEAN calculated zero signal + 3 SD : 0.5 – 1.5 pg/ml (shake – static incubation)
2x (MEAN calculated zero signal) : 1 – 3 pg/ml (shake – static incubation)

Expected values

IL-13 values in fresh serum and plasma samples of healthy individuals are below 10 pg/ml.

Specificity

No crossreactivity was observed with the following recombinant human proteins: IL-1α, IL-1β, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, Macrophage Colony Stimulating Factor (M-CSF), Granulocyte Colony Stimulating Factor (G-CSF), Granulocyte/Macrophage Colony Stimulating Factor (GM-CSF), Leukaemia Inhibitory Factor (LIF), RANTES, Stem Cell Factor/ Mast Cell Factor (SCF/MCF), Transforming Growth Factor β-1 (TGFβ-1), Tumour Necrosis Factor α, Tumour Necrosis Factor β (TNFβ/Lymphotoxin), and Interferon γ (IFNγ).

References

1. Mosmann, T.R. and Coffman, R.L. (1989) Annu.Rev.Immunol. 7: 145
2. Minty, A. *et al* (1993) Nature 362: 248
3. McKenzie, A.N.J. *et al* (1993) Proc.Natl.Acad.Sci. USA 90: 3735
4. Zurawski, G. and De Vries, J.E. (1994) Immunol.Today 15: 19
5. Zurawski, G. and De Vries, J.E. (1994) Stem Cells 12: 169
6. De Waal Malefyt, R. *et al* (1993) J.Immunol. 151: 6370
7. Defrance,T. *et al* (1994) J.Exp.Med. 179: 135
8. Punnonen, J. *et al* (1993) Proc.Natl.Acad.Sci. USA 90: 3730
9. Zurawski, G. *et al* (1993) EMBO J. 12: 2663
10. Jinquan,T.B. *et al* (1995) J.Immunol. 154: 3742
11. Van der Pouw Kraan, C.T.M. *et al* (1995) submitted
12. Labit-Le Bouteiller, C. *et al* (1995) J.Immunol.Meth. 181: 29

Reagents
Plesmanlaan 125
1066 CX Amsterdam
The Netherlands
Phone: +31 20 512 3599
Fax: +31 20 512 3570
E-mail: reagents@sanquin.nl
Website: www.sanquinreagents.com

p.t.o.

Standard

A natural human IL-13 standard has been calibrated against the WHO International Standard (IL-13 94/622; National Institute for Biological Standards and Control, Potters Bar, Hertfordshire, U.K. 1 WHO Unit = 1 ng IL-13).

The kit contains one lyophilized vial with natural IL-13.

Reconstitute the lyophilized standard by adding 500 µl of distilled water to the vial. Incubate for 10 minutes at room temperature and mix gently. After reconstitution the standard must be kept cold (2-8°C) and stored frozen after use (<-18°C, preferably <-70°C).

Standard curve

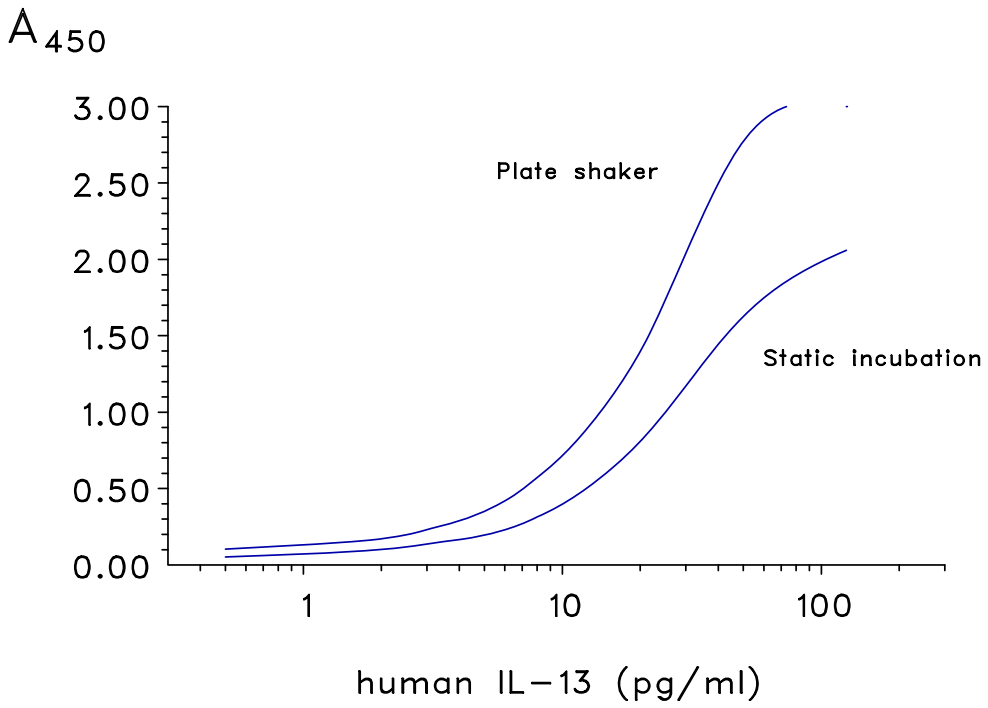
Label 7 tubes, one tube for each dilutions: 125, 50, 20, 8, 3.2, 1.3 and 0.5 pg/ml.
Pipette 420 µl of working-strength dilution buffer into the tube labelled 125 pg/ml and 300 µl of workingstrength dilution buffer into the other tubes.
Transfer 60 µl of the IL-13 standard (1000 pg/ml) into the first tube labelled 125 pg/ml, mix well and transfer 200 µl of this dilution into the second tube labelled 50 pg/ml.
Repeat the serial dilutions five more times by adding 200 µl of the previous tube of diluted standard to the 300 µl of dilution buffer.
The standard curve will contain 125, 50, 20, 8, 3.2, 1.3, 0.5 and 0 pg/ml (dilution buffer).

It is recommended to prepare two separate series for each assay.

Samples

It is recommended to dilute the test samples at least 1:2 in working-strength dilution buffer. If high levels of IL-13 (outside the standard curve) are expected in the test samples, additional dilutions of sample i.e. 1:20 and 1:100 should also be prepared.

Typical standard curve



	STATIC INCUBATION	SHAKEN INCUBATION
	Calculated mean absorbance at 450 nm	
substrate blank	0	0
0 pg/ml	0.036	0.065
0.5 pg/ml	0.053	0.104
1.3 pg/ml	0.081	0.144
3.2 pg/ml	0.158	0.289
8 pg/ml	0.314	0.573
20 pg/ml	0.808	1.394
50 pg/ml	1.627	2.772
125 pg/ml	2.030	≥ 3.000

DO NOT USE THESE DATA TO CONSTRUCT A STANDARD CURVE FOR SAMPLE VALUE CALCULATIONS