Human Interferon λ (IFN-λ) ELISA Kit

Cat#: E5054

Store at 2-8°C, 6 months

INTRODUCTION

This kit allows for the determination of IFN-λ concentrations in human serum, plasma, tissue homogenates and other biological fluids.

SPECIFICATION

Sensitivity: The minimum detectable dose is typically less than 1.0 pg/ml.

Detection ange: 31.25 pg/ml - 1000 pg/ml

PRINCIPLE OF KIT

The kit assay Human IFN-λ level in the sample, use purified Human IFN- λ antibody to coat microtiter plate wells, make solid-phase antibody, then add IFN-λ to the wells, combined antibody which With HRP labeled, become antibody-antigen-enzyme-antibody complex, after washing completely, add TMB substrate solution,TMB substrate becomes blue color at HRP enzyme-catalyzed, reaction is terminated by the addition of a sulphuric acid solution and the color change is measured spectrophotometrically at a wavelength of 450 nm. The concentration of IFN-λ in the samples is then determined by comparing the O.D. of the samples to the standard curve.

CONTENTS AND STORAGE

Components	48 Determinations	96 Determinations	Storage
User Manual	1	1	
Closure Plate Membrane	2	2	
Sealed Bags	1	1	
Microelisa Stripplate	1	1	2-8℃
Standard	0.3ml×6 bottle	0.3ml×6 bottle	2-8℃
HRP-Conjugate Reagent	5ml×1 bottle	10ml×1 bottle	2-8℃
Sample Diluent	3ml×1 bottle	6ml×1 bottle	2-8℃
Chromogen Solution A	3ml×1 bottle	6ml×1 bottle	2-8℃
Chromogen Solution B	3ml×1 bottle	6ml×1 bottle	2-8℃
Stop Solution	3ml×1 bottle	6ml×1 bottle	2-8℃
20×Wash Solution	15ml×1 bottle	25ml×1 bottle	2-8℃

Note: Standard concentration was followed by: 1000、500、250、125、62.5、0 pg/ml.

ACE BIOLABS CO., LTD.   www.acebiolab.com

6F., No 68, Qingsheng 2nd St., Dayuan Dist., Taoyuan City33743, Taiwan.

NOTES

1. The kit takes out from the refrigeration environment should be balanced 15-30 minutes in the room temperature, ELISA plates coated if has not use up after opened, the plate should be stored in Sealed bag.

2. Washing buffer will crystallization separation, it can be heated the water helps dissolve when dilute . Washing does not affect the result.

3. Add sample with sampler each step, and proofread its accuracy frequently, avoids the experimental error. add sample within 5 mins, if the number of sample is much, recommend to use Volley .

4. If the testing material content is excessively higher (The sample OD is bigger than the first standard well), please dilute Sample (n-fold), please diluente and multiplied by the dilution factor.(×n×5).

5. Closure plate membrane only limits the disposable use, to avoid cross-contamination.

6. The substrate evade the light preservation.

7. Please according to use instruction strictly, the test result determination must take the microtiter plate reader as a standard.

8. All samples, washing buffer and each kind of reject should according to infective material process.

9. Do not mix reagents with those from other lots.

SAMPLE COLLECTION

1. **Serum**-coagulation at room temperature 10-20mins, centrifugation 20-min at the speed of 2000-3000 r.p.m. remove supernatant, if precipitation appeared, centrifugal again.

2. **Plasma**-use suited EDTA or citrate plasma as an anticoagulant, mix 10-20 mins ,centrifugation 20-min at the speed of 2000-3000 r.p.m. remove supernatant, if precipitation appeared, centrifugal again.

3. **Urine**-collect sue a sterile container, centrifugation 20-min at the speed of 2000-3000 r.p.m. remove supernatant, if precipitation appeared, centrifugal again. The operation of hydrothorax and cerebrospinal fluid reference to it.

4. **Cell culture supernatant**-detect secretory components, collect sue a sterile container, centrifugation 20-min at the speed of 2000-3000 r.p.m. remove supernatant, detect the composition of cells, dilut cell suspension with PBS (pH7.2-7.4), cell concentration reached 1 million / ml, repeated freeze-thaw cycles, damage cells and release of intracellular components, centrifugation 20-min at the speed of 2000-3000 r.p.m. remove supernatant, if precipitation appeared, Centrifugal again.

5. **Tissue samples**- After cutting samples, check the weight,add PBS (pH7.2-7.4), rapidly frozen with liquid nitrogen, maintain samples at 2-8°C after melting,add PBS (pH7.4), homogenized by hand or grinders, centrifugation 20-min at the speed of 2000-3000 r.p.m. remove supernatant.

6. Extract as soon as possible after specimen collection, and according to the relevant literature,

and should be experiment as soon as possible after the extraction. If it can't, specimen can be kept in -20 °C to preserve, avoid repeated freeze-thaw cycles.

7. Can't detect the sample which contain NaN_3, because NaN_3 inhibits HRP active.

ASSAY PROCEDURE

1. Add standard: Set Standard wells, testing sample wells. Add standard 50µl to standard well.

2. Add sample: Set blank wells separately (blank comparison wells don't add sample and HRP-conjugate reagent, other each step operation is same). Testing sample well. add sample dilution 40µl to testing sample well, then add testing sample 10µl (sample final dilution is 5-fold), add sample to wells , don't touch the well wall as far as possible, and gently mix.

3. Add enzyme: Add HRP-Conjugate reagent 100µl to each well, except blank well.

4. Incubate: After closing plate with closure plate membrane ,incubate for 60 min at 37°C.

5. Configure liquid: 20-fold wash solution diluted 20-fold with distilled water and reserve.

6. Washing: Uncover closure plate membrane, discard liquid, dry by swing, add washing buffer to every well, still for 30s then drain, repeat 5 times, dry by pat.

7. Color: Add Chromogen Solution A 50ul and Chromogen Solution B to each well, evade the light preservation for 15 min at 37°C

8. Stop the reaction: Add Stop Solution 50µl to each well, stop the reaction(the blue color change to yellow color).

9. Assay: Take blank well as zero , read absorbance at 450nm after adding stop Solution and within 15min.

CALCULATION Of RESULTS

Take the standard density as the horizontal, the OD value for the vertical, draw the standard curve on graph paper, find out the corresponding density according to the sample OD value by the sample curve, multiplied by the dilution multiple, or calculate the straight line regression equation of the standard curve with the standard density and the OD value, with the sample OD value in the equation, calculate the sample density.

standards concentration (X)

ACE
Biolabs
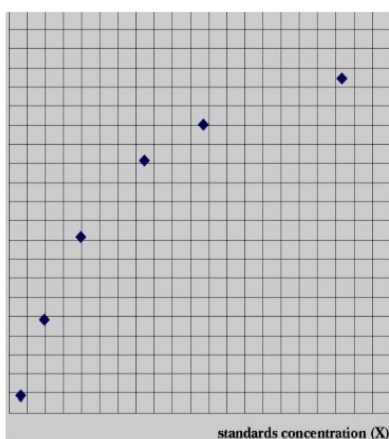
DECLARATION

1. Limited by current conditions and scientific technology, we can't conduct comprehensive identification and analysis on all the raw material provided. So there might be some qualitative and technical risks for users using the kit.

2. The final experimental results will be closely related to the validity of products, operational skills of the operators and the experimental environments. Please make sure that sufficient samples are available.

3. To get the best results, please only use the reagents supplied by the manufacturer and strictly comply with the instructions!

4. Incorrect results may occur because of incorrect operations during the reagents preparation and loading, as well as incorrect parameter settings of the Micro-plate reader. Please read the instructions carefully and adjust the instrument prior to the experiment.

5. Even the same operator might get different results in two separate experiments. In order to get reproducible results, the operation of every step in the assay should be controlled.

6. Every kit has strictly passed QC test. However, results from end users might be inconsistent with our data due to some variables such as transportation conditions, different lab equipments, and so on. Intra-assay variance among kits from different batches might arise from the above reasons, too.