Instructions for Use of Human IFN-γ ELISA Detection Kit

This kit is intended for scientific use and not for diagnostic use

Cat. No. HG-IF001

Product introduction

Human IFN-γ ELISA Detection Kit in BlueKit® series uses a double-antibody sandwich method for quantitative detection of human IFN-γ protein content in serum, plasma or cell supernatant. Coat the specific anti-human IFN-γ monoclonal antibody on a microplate, add the standard , test sample, and detection antibody into the reaction wells, incubate at room temperature (18-25℃), wash, and then add the Streptavidin-HRP for incubation. After washing, add chromogenic solution TMB. The shade of the color is proportional to the target protein concentration.

Detection range: 1.37-1000pg/mL

Sensitivity: 0.36 pg/mL

Precision: CV% ≤ 10%, RE% ≤ ±15%

Specification

96T

Usage

The product is used for the assay of human IFN-γ protein content in serum, plasma, cell culture supernatant and other biological samples.

Kit composition

Components	Specification	Preparation
Coated microtiter plate	8 wells x 12 strips	Ready-to-use
Human IFN-γ Standards	S, S1 - S7, S0	Ready-to-use
Detection Antibody (DA)	6 mL × 1 bottle	Ready-to-use
Streptavidin-HRP (SH)	12 mL × 1 bottle	Ready-to-use
1x Detection Buffer (AB)	12 mL × 1 bottle	Ready-to-use
10x Wash Concentrate	50 mL × 1 bottle	Dilute with deionized water in a ratio of 1:9
Chromogenic solution	12 mL × 1 bottle	Ready-to-use
Stop solution	12 mL × 1 bottle	Ready-to-use
Sealing film	3 films	
Instructions for Use	1 copy	

Notes: All components are stored at 2-8℃.

Storage conditions and shelf life

The unopened kit is valid for 12 months at 2-8℃.

Materials to be self-prepared:

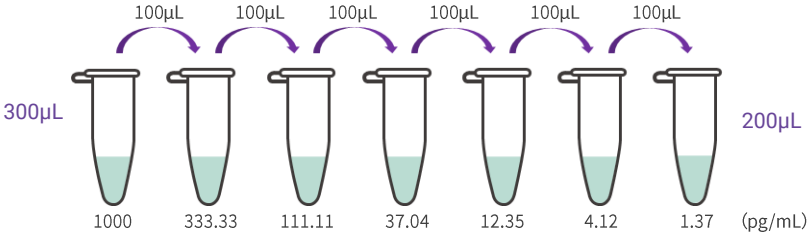
(1) Microplate reader
(2) Constant temperature incubator
(3) Micropipettes and tips
(4) Deionized or distilled water
(5) New filter paper
(6) Vortex oscillator

Reagent configuration

1x wash solution: According to the actual amount, take an appropriate amount of 10x wash concentrate and dilute it by 10 times with deionized water.

Creation of a standard curve

S1 to S7 and S0 are used directly for serum and plasma sample detection.
For other sample types, create a standard curve with sample preparation buffer (SPB), e.g. cell culture supernatant, tissue grind, cell lysate, etc. Urine samples are used to draw a standard curve using AB (detection buffer).
Human IFN-γ Standard S 10000 pg/mL 30 μL+270 μL SPB serves as the high standard (1000 pg/mL). 200 μL of SPB is added to each dilution tube and 1:2 dilution series are prepared using the high standard. Thoroughly mix each tube before performing the next transfer. SPB is used as a zero standard (0 pg/mL).



Operating steps

Restore all reagents and samples to room temperature (18-25℃) before testing.
(1) Prepare all required reagents and working concentration standards.
(2) Remove the unwanted strips, place them back into the foil pouch, and seal them again.
(3) Add 50 μL detection buffer (AB) to each well.
(4) Add 50 μL of standard (S) and sample. Ensure continuous spiking without interruption. The spiking process shall be completed within 15 minutes.

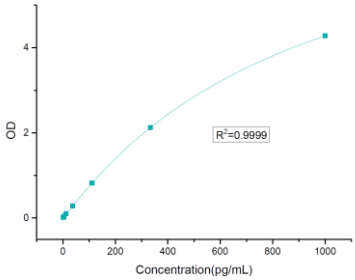
(5) Add 50 µL of detection antibody (DA) to each well.
(6) Seal the plate with a sealing film. Shake at 500 rpm and incubate at room temperature (18-25℃) for 30 minutes.
(7) Discard the liquid in the wells, add 300 µL wash solution to each well to wash the plate, and wash 4 times. Each time you wash the plate, pat it dry on absorbent tissue. For ideal experimental performance, the residual liquid must be removed thoroughly.
(8) Add 100 µL Streptavidin-HRP (SH).
(9) Seal the plate with a new sealing film. Shake at 500 rpm and incubate at room temperature (18-25℃) for 15 minutes.
(10) Repeat step 7.
(11) Add 100 µL of chromogenic solution to each well and incubate at room temperature (18-25℃) for 10-15 minutes.
(12) Add 100 µL stop solution to each well.
(13) Within 30 minutes, determine the OD value at 450 nm wavelength of the microtiter plate, and set the correction wavelength as 570 nm or 630 nm.

Result processing

(1) OD processing of the standard curve (See the following example. For example only, please refer to the actual measurement for details)

Standard concentration (pg/mL)	OD1	OD2	Mean value
1000.00	4.1920	4.3700	4.2810
333.33	2.0721	2.1701	2.1211
111.11	0.8112	0.8352	0.8232
37.04	0.2915	0.2705	0.2810
12.35	0.1072	0.0989	0.1031
4.12	0.0382	0.0340	0.0361
1.37	0.0124	0.0160	0.0142
0.00	0.0098	0.0106	0.0102

(2) The standard curve is obtained by 4-parameter fitting with the theoretical standard concentrations and the corresponding OD values (as shown in the figure below).



Precautions

(1) For the first detection of samples, it is recommended to perform at least three consecutive dilutions to produce at least one diluted sample within the range of the standard curve.

(2) Store reagents according to label instructions and equilibrate at room temperature (18-25℃) before use.

(3) Before using the coated microtiter plate, please balance to room temperature (18-25℃) and then open the secondary package. The strips not used in the experiment shall be immediately placed back in the package for sealing and can be stored at 4℃ for one month. The remaining reagents shall be packaged or covered.

(4) Please use disposable tips during the experimental operation to avoid cross-contamination.

(5) Check various reagents in the kit before use. Dilution, spiking and termination of the reaction with reagents shall be thoroughly mixed or shaken well, which is particularly important for the experimental results.

(6) The residual wash solution in the reaction wells during the washing process shall be patted thoroughly on a clean tissue until no watermark is visible. Do not place the tissue directly into the reaction wells to absorb water.

(7) The substrate chromogenic solution is sensitive to light. Avoid prolonged exposure to light and avoid contact with metals that may affect the results.

(8) This product is a disposable kit and shall be used within the validity period.

Disclaimer

Under all circumstances, the liability of our company for this product is only limited to the value of the product itself.

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