PRODUCT INFORMATION

Human IL-10 Matched Antibody Pair

v. 240101

Catalog number	C10045-bulk / C10045-K01
Package	Customized package / Set
Product Description	The Human IL-10 Antibody Pair Kit includes pre-matched antibody pairs and standards, allowing for the development of customized enzyme-linked immunosorbent assays (ELISA) to detect and quantify human IL-10 protein levels. The kit comprises an unlabeled capture antibody, an HRP-conjugated detection antibody, and a standard protein. This Antibody Pair Kit, featuring mouse monoclonal antibodies, has the potential for quantifying both natural and recombinant human Interleukin-10 (IL-10) in ELISA and other immunoassays.

Components

Reagents	Quantity	Form
Human IL-10 Capture Antibody	350 µL, 1 vial	Liquid
Human IL-10 Detection Antibody (HRP conjugated)	350 µL, 1 vial	Liquid
Human IL-10 Standard	125 ng/vial, 2 vials	Lyophilized

** The kit provides raw materials for around 96 tests across 5 plates. However, actual results may vary based on the researcher's assay protocol and other variables.*

Recommended Dilution and Reconstitution

Human IL-10 Capture Antibody
Dilute 200X with PBS to the working concentration and add 100 µL/well.

Human IL-10 Detection Antibody
Dilute 200X with PBS to the working concentration and add 100 µL/well.

Human IL-10 Standard
Refer to the lot-specific CoA for reconstitution volume. Reconstitute each vial with 1% BSA in PBS. Standard curve using 2-fold serial dilutions is recommended.

Storage and Stability

Check the box front label for the expiration date.

- Before opening or reconstituting, store all kit reagents properly at 2-8°C .
- Once opened, use the kit immediately. Return remaining reagents to 2-8°C after use, except the reconstituted standard, which must be stored at -80°C.
- Avoid multiple freeze-thaw cycles for the frozen reconstituted standard. If stored properly at -80°C, it is valid for a maximum of 2 weeks.
- Freshly prepare both the Capture Antibody and Detection Antibody in their working concentration and use them on the same day.

Materials Required but not Provided	**Devices & Consumables** 1. 10 mL graduated pipettes 2. 5 µL to 1000 µL adjustable single-channel micropipettes with disposable tips 3. 50 µL to 300 µL adjustable multi-channel micropipettes with disposable tips 4. Multi-channel micropipette reservoir 5. Disposable microcentrifuge tubes 6. Beakers, flasks, cylinders necessary for preparation of reagents 7. Timer 8. Magnetic stirrer 9. Vortex mixer 10. Washer for microplates 11. Incubator capable of maintaining temperature at 37±1°C 12. Stripwell microplate spectrophotometer capable of reading at 450 nm 13. Clean paper towels 14. Disposable gloves 15. Discard container for bio-medical waste 16. 96-well ELISA plate 17. Microplate sealing film **Reagents** 1. High quality distilled water 2. Standard & Sample diluent buffer 3. Wash buffer 4. TMB 5. Stop solution
Intended Use	The Human IL-10 Matched Antibody Pair facilitates the quantitative determination of Human Interleukin-10 through a sandwich ELISA. This antibody pair includes essential components necessary for the development of the ELISA assay, streamlining the process for researchers aiming to measure Human IL-10 levels accurately.

General Assay Procedure

A. Evaluate the number of stripwell required to test the samples. Put the stripwells at room temperature (15-30°C) before use. The unused strips should be resealed in the bag and stored at 2-8°C. Each standard, blank, and sample should be assayed in duplicate.

B. Standard and sample preparation:

Standard preparation (in microcentrifuge tubes)

1. Refer to the vial label for reconstitution volume. Reconstitute the lyophilized standard with Standard reconstitution buffer to the concentration of 125 ng/ mL.
2. Vortex for 1 min and incubate for at least 10 minutes. Aliquot and store the standards at -20°C.
3. Add 400 µL Standard & Sample diluent buffer to 100 µL of 125 ng/ mL standard to make a 25 ng/ mL standard (Tube 1).
4. Adding 250 µL of Standard & Sample diluent buffer to 250 µL of 25 ng/ mL standard to make a 12.5 ng/ mL standard (Tube 2).
5. Repeat the above procedure to make serial diluted standards (Tube 3-7).
6. Tube 8 is blank which only contains Standard & Sample diluent buffer.

Sample preparation:

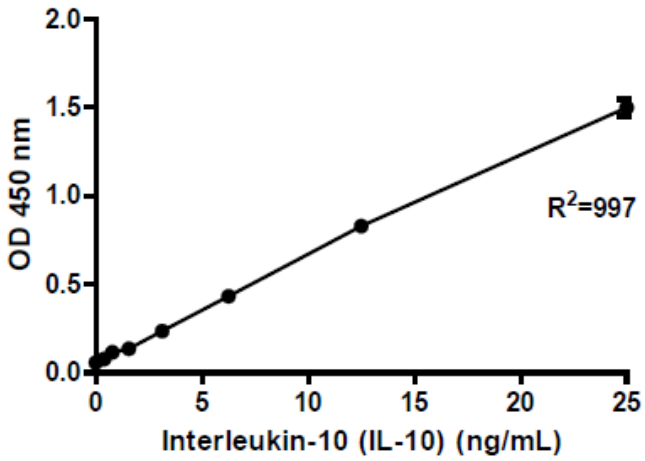
1. 100 µL Sample. If the initial assay found samples contain Interleukin-10 (IL-10) higher than the highest standard, the samples can be diluted with Standard & Sample diluent buffer and then re-assay the samples.
2. Add 100 µL of standards, blanks or samples into Interleukin-10 (IL-10) ELISA stripwell microplates (see Table 1), then add 100 µL of Assay buffer into each well immediately. Cover with microplate sealing film and incubate sealed plate at 37°C for 2 hour.
3. Remove the sealing film, aspirate the liquid from each well and then wash the plate three times with 300 µL Working wash buffer per well. After the last wash, tap stripwells on clean absorbent paper to remove excess wash buffer.
4. Add 100 µL of Working HRP-antibody conjugate into each well. Cover with microplate sealing film and incubate sealed plate at 37℃ for 1 hour in the dark.
5. Remove the sealing film, aspirate the liquid from each well and then wash the plate six times with 300 µL Working wash buffer per well. After the last wash, tap stripwells on clean absorbent paper to remove excess wash buffer.
6. Add 100 µL of TMB into each well. Incubate for 10 minutes at room temperature (15-30°C) in the dark.
7. Add 50 µL Stop solution into each well.
8. Read the absorbencies immediately at 450 nm after the Stop solution is added.

Table1. An example of orientation of standards, blanks and samples in the stripwells microplate**.**

	1	2	3	4
A	Standard 1 (25 ng/mL)	Standard 1 (25 ng/mL)	Sample 1	Sample 5
B	Standard 2 (12.5 ng/mL)	Standard 2 (12.5 ng/mL)	Sample 1	Sample 5
C	Standard 3 (6.25 ng/mL)	Standard 3 (6.25 ng/mL)	Sample 2	Sample 6
D	Standard 4 (3.125 ng/mL)	Standard 4 (3.125 ng/mL)	Sample 2	Sample 6
E	Standard 5 (1.563 ng/mL)	Standard 5 (1.563 ng/mL)	Sample 3	Sample 7
F	Standard 6 (0.781 ng/mL)	Standard 6 (0.781 ng/mL)	Sample 3	Sample 7
G	Standard 7 (0.391 ng/mL)	Standard 7 (0.391 ng/mL)	Sample 4	Sample 8
H	Blank	Blank	Sample 4	Sample 8

Typical data

The presented standard curve is for demonstration purposes only. It is essential to generate a standard curve for each individual assay.



For Research Use Only.

Precautions & Warnings

In order to obtain reproducible test results, the following rules should be strictly obeyed:

- All reagents and specimens should be considered as potentially hazardous. We therefore recommend that this product is handled by those persons who have been properly trained.
- Wear suitable protective clothing and disposable gloves.
- Care should be taken to avoid reagents contacting with skin or eyes. If contacted, wash immediately and thoroughly with plenty of clean water.
- This product is intended for Research use only and is not for use in diagnostic and therapeutic procedures.
- This product is designed for a single, one-time use only.
- The assay should be performed as outlined in this manual, and in accordance with all instructions.
- Do not use expired or damaged products.
- Do not mix or substitute reagents with those from different lots or other sources.
- Thoroughly and gently mix all the reagents and specimens prior to use.
- Do not expose all the reagents to strong light during storage or incubation.
- Use disposable graduated pipettes and tips to avoid microbial contamination or cross-contamination of reagents or specimens which may invalidate the test.
- After use, all the reagents and specimens should be regarded as medical waste with risk of biological infection and properly disposed of in accordance with national regulations.