ELISA Buffer Set-96T

Pack Size: 96 tests

Catalog Number： EBS-001

IMPORTANT: Please carefully read this manual before performing your experiment.

For Research Use Only. Not For Use In Diagnostic Or Therapeutic Procedure

MATERIALS PROVIDED

TABLE 1. MATERIALS PROVIDED

ID	Composition	Size	Detail	Storage
P001	High-bind plate	1 plate	96 well	2-8℃
B001	Coating buffer	12 mL	1 X	2-8℃
B002	Washing buffer	60 mL	10 X	2-8℃
B003	Blocking buffer	5 mL	10 X	2-8℃
B004	Substrate solution	12 mL	1 X	2-8℃, avoid light
B005	Stop Solution	7 mL	1 X	2-8℃

COMPOSITION

This ELISA Buffer Set-96T contains a High-bind 96well plate which contains 96 wells, flat bottom, without lid, high binding EIA/RIA plate.

Coating buffer contains $NaHCO_3$, Na_2CO_3, pH 9.6.

Washing buffer contains Tris, NaCl, Tween20, pH7.4.

Blocking buffer contains BSA in Washing Buffer, pH7.4.

Substrate solution contains TMB in Substrate Solution ($Na_2HPO_4 \cdot 12H_2O$, Citric acid, pH5.5).

Stop Solution contains 1 mol/L sulfuric acid.

The ELISA Buffer Set-96T Can be quickly applied to the ELISA experiment and save your time to prepare buffers. This ELISA Buffer Set-96T is sterile-filtered.

Note: *Substrate solution should be protected from light.*

SHIPPING

This ELISA Buffer Set-96T is very stable, even at room temperature. It is recommended to ship this product at room temperature.

STORAGE

Store this set at 2-8℃, and protect it from light. If precipitation occurs, filter to clarify before use.

This set is stable after storage at 2-8℃ no less than 3 months upon receipt.

Note: *All items recover to room temperature before experiment.*

APPLICATION

Coating buffer: Use coating buffer to dilute the sample you will coat, add 100μL/well to high-bind plate.

Washing buffer: Dilute the washing buffer to 1x washing buffer with distilled water (For example, 60mL 10 x washing buffer + 540 mL distilled water) before experiment, add 300μL/well for washing the coated plate, wash the plate for 3 times.

Blocking buffer: Dilute the 10 x blocking buffer to 1x with distilled water (For example, 5mL 10 x blocking buffer + 45 mL distilled water) before experiment, add 300μL/well for blocking the coated plate.

Sample buffer: Dilute the 1x blocking buffer with 3 times volume washing buffer to make sample/detection antibody buffer (For example, 1mL 1 x blocking buffer + 3 mL 1 x washing buffer) before experiment.

Substrate solution: Add 100μL/well to plate.

Stop solution: Add 50μL/well to plate.

TYPICAL DATA

1. **Coating**
   Coat the plate (ID. P001) with 0.2 μg/well (2 μg/ml, 100 μl/well) Human IL-15, Tag Free (Cat. No. IL5-H4117, ACRO Biosystems) at 4 ℃ for overnight (or 16 hours). The protein is diluted in Coating Buffer (ID. B001).
2. **Washing**
   Wash the wells with 300μL per well Washing Buffer (ID. B002, 1x) for 4 times. Please note that the complete removal of the Washing buffer is essential. After washing, remove remaining solution by aspirating or decanting. Invert the plate and let it sit on clean paper towels for a while to make sure it's completely dried.
3. **Blocking**
   Block the wells with 300μL Blocking Buffer (ID. B003, 1x) per well at 37 ℃ for 1.5 hours.
4. **Washing**
   Repeat step 2.
5. **Adding Sample**

Add 100µl 0.610352-78.125 ng/mL Human IL-15 R alpha Fc Chimera Protein to each well, and incubate at 37 ℃ for 1 hour. The sample is diluted in Sample Buffer (1x blocking buffer with 3 times volume washing buffer).

6. **Washing**
   Repeat step 2.

7. **Adding Detection Antibody**

   Add 100µL Peroxidase AffiniPure Goat Anti-Human IgG, Fcγ fragment specific (min X Bov, Hrs, Ms Sr Prot) (Jackson, Cat. No. 109-035-098) to each well, and incubate at 37 ℃ for 1 hour. The antibody is diluted 1:20000 in Sample Buffer (1x blocking buffer with 3 times volume washing buffer).

8. **Washing**
   Repeat step 2

9. **Adding Substrate**

   Add 100µL Substrate Solution (ID. B004) into each well, incubate at 37 ℃ for 20 min. Avoid light.

10. **Termination**
    Add 50µL 1 mol/L Stop Solution (ID. B005) to each well.

11. **ReadOD**
    Read OD at 450 nm, then OD450-Blank is the final OD Value.

12. **Data Analysis**

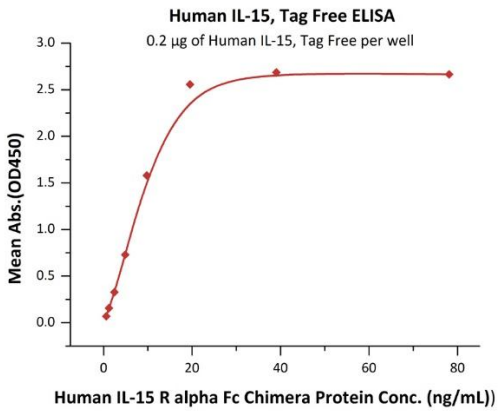

Fig1. Immobilized Human IL-15, Tag Free (SPR verified) (Cat. No. IL5-H4117) at 2 µg/mL (100 µL/well) can bind Human IL-15 R alpha Fc Chimera Protein with a linear range of 0.6-20 ng/mL.