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CCL25 ELISA Kit





Overview

Quantity:	96 tests
Target:	CCL25
Reactivity:	Mouse
Method Type:	Sandwich ELISA
Detection Range:	15-2000 pg/mL
Minimum Detection Limit:	15 pg/mL
Application:	ELISA

Product Details

Product Details	
Purpose:	Mouse TECK (CCL25) ELISA Kit for cell culture supernatants, plasma, and serum samples.
Sample Type:	Plasma, Cell Culture Supernatant, Serum
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This ELISA kit shows no cross-reactivity with any of the cytokines tested: Mouse 6Ckine, CTACK, Eotaxin, GCSF, GM-CSF, IL-2, IL-3, IL-4, IL-5, IL-6, IL-9, IL-10, IL-12p40, IL-12p70, IL-13, IL-17, IFN-gamma, KC, Leptin, MCP-5, MIP-1 alpha, MIP-2, MIP-3 beta, RANTES, SCF, sTNFri, TARC, TIMP-1, TNF-alpha, TPO, VEGF.
Sensitivity:	15 pg/mL
Characteristics:	 Strip plates and additional reagents allow for use in multiple experiments Quantitative protein detection Establishes normal range

Product Details

	The best products for confirmation of antibody array data
Components:	Pre-Coated 96-well Strip Microplate
	Wash Buffer
	Stop Solution
	Assay Diluent(s)
	Lyophilized Standard
	Biotinylated Detection Antibody
	Streptavidin-Conjugated HRP
	TMB One-Step Substrate
Material not included:	Distilled or deionized water
	 Precision pipettes to deliver 2 μL to 1 μL volumes
	 Adjustable 1-25 μL pipettes for reagent preparation
	• 100 µL and 1 liter graduated cylinders
	Tubes to prepare standard and sample dilutions
	Absorbent paper
	Microplate reader capable of measuring absorbance at 450nm
	 Log-log graph paper or computer and software for ELISA data analysis

Target Details

Target:	CCL25
Alternative Name:	TECK (CCL25 Products)
Background:	C-C motif chemokine 25 (Chemokine TECK) (Small-inducible cytokine A25) (Thymus-expressed chemokine)
Gene ID:	20300
UniProt:	035903

Application Details

Application Notes:	Recommended Dilution for serum and plasma samples2 fold
Sample Volume:	100 μL
Plate:	Pre-coated
Protocol:	 Prepare all reagents, samples and standards as instructed in the manual. Add 100 μL of standard or sample to each well. Incubate 2.5 h at RT or O/N at 4 °C. Add 100 μL of prepared biotin antibody to each well.

- 5. Incubate 1 h at RT.
- 6. Add 100 µL of prepared Streptavidin solution to each well.
- 7. Incubate 45 min at RT.
- 8. Add 100 µL of TMB One-Step Substrate Reagent to each well.
- 9. Incubate 30 min at RT.
- 10. Add 50 µL of Stop Solution to each well.
- 11. Read at 450 nm immediately.

Reagent Preparation:

1. Bring all reagents and samples to room temperature (18 - 25°C) before use. 2. Sample dilution: If your samples need to be diluted, Assay Diluent A (Item D) is used for dilution of serum/plasma samples, and Assay Diluent B (Item E) is used for dilution of culture supernatants. 3. Assay Diluent B should be diluted 5-fold with deionized or distilled water. 4. Preparation of standard: Briefly spin the vial of Item C and then add 400 µl Assay Diluent A (for serum/plasma samples) or 1x Assay Diluent B (for cell culture medium) into Item C vial to prepare a 50 ng/ml standard. Dissolve the powder thoroughly by a gentle mix. Add 40 µl TECK standard from the vial of Item C, into a tube with 960 µl Assay Diluent A or 1x Assay Diluent B to prepare a 2,000 pg/ml stock standard solution. Pipette 300 µl Assay Diluent A or 1x Assay Diluent B into each tube. Use the stock standard solution to produce a dilution series (shown below). Mix each tube thoroughly before the next transfer. Gently vortex to mix. Assay Diluent A or 1x Assay Diluent B serves as the zero standard (0 pg/ml). 5. If the Wash Concentrate (20x) (Item B) contains visible crystals, warm to room temperature and mix gently until dissolved. Dilute 20 ml of Wash Buffer Concentrate into deionized or distilled water to yield 400 ml of 1x Wash Buffer. 6. Briefly spin the Detection Antibody vial (Item F) before use. Add $100 \, \mu l$ of 1xAssay Diluent B into the vial to prepare a detection antibody concentrate. Pipette up and down to mix gently (the concentrate can be stored at 4°C for 5 days). The detection antibody concentrate should be diluted 65-fold with 1x Assay Diluent B and used in step 4 of Part VI Assay Procedure. 7. Briefly spin the HRP-Streptavidin concentrate vial (Item G) and pipette up and down to mix gently before use. HRP-Streptavidin concentrate should be diluted 6,000-fold with 1x Assay Diluent B. For example: Briefly spin the vial (Item G) and pipette up and down to mix gently . Add 2 µl of HRP-Streptavidin concentrate into a tube with 12 ml 1x Assay Diluent B to prepare 6,000 fold diluted HRP- Streptavidin solution.

Assay Procedure:

1. Bring all reagents and samples to room temperature ($18 - 25^{\circ}$ C) before use. It is recommended that all standards and samples be run at least in duplicate. 2. Add $100 \, \mu l$ of each standard (see Reagent Preparation step 2) and sample into appropriate wells. Cover well and incubate for 2.5 hours at room temperature or over night at 4°C with gentle shaking. 3. Discard the solution and wash 4 times with 1x Wash Solution. Wash by filling each well with Wash Buffer ($300 \, \mu l$) using a multi-channel Pipette or autowasher. Complete removal of liquid at each

step is essential to good performance. After the last wash, remove any remaining Wash Buffer by aspirating or decanting. Invert the plate and blot it against clean paper towels. 4. Add 100 μ l of 1x prepared biotinylated antibody (Reagent Preparation step 6) to each well. Incubate for 1 hour at room temperature with gentle shaking. 5. Discard the solution. Repeat the wash as in step 3. 6. Add 100 μ l of prepared Streptavidin solution (see Reagent Preparation step 7) to each well. Incubate for 45 minutes at room temperature with gentle shaking. 7. Discard the solution. Repeat the wash as in step 3. 8. Add 100 μ l of TMB One-Step Substrate Reagent (Item H) to each well. Incubate for 30 minutes at room temperature in the dark with gentle shaking. 9. Add 50 μ l of Stop Solution (Item I) to each well. Read at 450 nm immediately.

Calculation of Results:

Calculate the mean absorbance for each set of duplicate standards, controls and samples, and subtract the average zero standard optical density. Plot the standard curve on log-log graph paper or using Sigma plot software, with standard concentration on the x-axis and absorbance on the y-axis. Draw the best-fit straight line through the standard points.

Restrictions:

For Research Use only

Handling

Storage: -20 °C

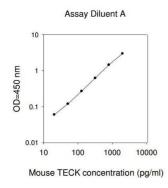
Storage Comment:

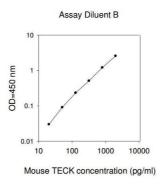
The entire kit may be stored at -20°C for up to 1 year from the date of shipment. Avoid repeated freeze-thaw cycles. The kit may be stored at 4°C for up to 6 months. For extended storage, it is recommended to store at -80°C.

Expiry Date:

6 months

Images





ELISA

Image 1.