Datasheet for ABIN625087

ICAM1 ELISA Kit

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Overview

Quantity:	96 tests
Target:	ICAM1
Reactivity:	Human
Method Type:	Sandwich ELISA
Detection Range:	150 pg/mL-20 ng/mL
Minimum Detection Limit:	150 pg/mL
Application:	ELISA

Product Details

Purpose:	Human ICAM-1 (CD54) 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: Human Angiogenin, BDNF, BLC, ENA-78, FGF-4, IL-1 alpha, IL-1 beta, IL-2, IL-3, IL-4, IL-5, IL-7, IL-8, IL-9, IL-10, IL-11, IL-12 p70, IL-12 p40, IL-13, IL-15, I-309, IP-10, G-CSF, GM-CSF, IFN-gamma, Leptin, MCP-1, MCP-2, MCP-3, MDC, MIP-1 alpha, MIP-1 beta, MIP-1 delta, PARC
Sensitivity:	150 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:	ICAM1
Alternative Name:	ICAM-1 (ICAM1 Products)
Target Type:	Viral Protein
Background:	SICAM-1 (soluble Intercellular Adhesion Molecular-1) has been reported in serum, cerebrospinal fluid and bronchoalveolar lavage. ICAM-1 expression is weak on leukocytes, epithelial and resting endothelial cells. ICAM-1 is a ligand for LFA-1 (CD11a/CD18) and Mac-1 (CD11b/CD18). Its expression is up-regulated upon stimulation by inflammatory mediators such as cytokines and LPS. The Human sICAM-1 ELISA (Enzyme-Linked Immunosorbent Assay) kit is an in vitro enzyme-linked immunosorbent assay for the quantitative measurement of human soluble ICAM-1 in serum, plasma, cell culture supernatants and urine. This assay employs an antibody specific for human sICAM-1 coated on a 96-well plate. Standards and samples are pipetted into the wells and sICAM-1 present in a sample is bound to the wells by the immobilized antibody. The wells are washed and biotinylated anti-human sICAM-1 antibody is added. After washing away unbound biotinylated antibody, HRP-conjugated streptavidin is pipetted to the wells. The wells are again washed, a TMB substrate solution is added to the wells and color develops in proportion to the amount of sICAM-1 bound. The Stop Solution changes the color from blue to

Target Details

	yellow, and the intensity of the color is measured at 450 nm. Reproducibility: Intra-Assay: CV<10% Inter-Assay: CV<12%.
Gene ID:	3383
UniProt:	P05362
Pathways:	Cellular Response to Molecule of Bacterial Origin, Regulation of Actin Filament Polymerization, Carbohydrate Homeostasis, Regulation of Leukocyte Mediated Immunity, Thromboxane A2 Receptor Signaling

Application Details

Application Notes:	Recommended Dilution for serum and plasma samples5 - 200 fold
Sample Volume:	100 µL
Plate:	Pre-coated
Protocol:	1. Prepare all reagents, samples and standards as instructed in the manual. 2. Add 100 µL of standard or sample to each well. 3. Incubate 2.5 h at RT or O/N at 4 °C. 4. 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) should be used for dilution of serum/plasma samples. Assay Diluent B (Item E) should be used for dilution of culture supernatants and urine. Suggested dilution for normal serum/plasma: 5-200 fold*. * Please note that levels of the target protein may vary between different specimens. Optimal dilution factors for each sample must be determined by the investigator. 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 750 µL Assay Diluent A (for serum/plasma samples) or 1x Assay Diluent B (for cell culture medium and urine) into Item C vial to prepare a 20 ng/mL standard. Dissolve the powder thoroughly by a gentle mix. Pipette 250 µL Assay Diluent A or 1x Assay Diluent B into each tube. Use the 20 ng/mL standard

	solution to produce a dilution series . Mix each tube thoroughly before the next transfer. Assay Diluent A or 1x Assay Diluent B serves as the zero standard (0 ng/mL). The 20 ng/mL standard in Assay Diluent B may be saturated, we recommend 10 ng/mL serves as starting point (the highest standard point) for Assay Diluent B. 250 µL Standard, Item C + 750 µL 250myl 250 µL 250 µL 250 µL 250 µL 20 10 5 2.5 1.25 0.625 0.313 0 ng/mL ng/mL ng/mL ng/mL ng/mL ng/mL ng/mL ng/mL ng/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 µL of 1x Assay 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 80-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 340-fold with 1x Assay Diluent B. For example: Briefly spin the vial (Item G) and pipette up and down to mix gently . Add 25 µL of HRP-Streptavidin concentrate into a tube with 8.5 ml 1x Assay Diluent B to prepare a 340-fold diluted HRP-Streptavidin solution (don't store the diluted solution for next day use). Mix well.
Assay Procedure:	1. Bring all reagents and samples to room temperature (18 - 25 °C) before use. It is recommended that all standards and samples be run at least in duplicate. 2. Add 100 µL 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 myl) 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 µL 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 6. Add 100 µL 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 8. Add 100 µL of TMB One-Step Substrate Reagent (Item H) to each well. Incubate for 30

Application Details

	minutes at room temperature in the dark with gentle shaking. 9. Add 50 µL 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. Typical Data: These standard curves are for demonstration only. A standard curve must be run with each assay. Assay Diluent A Human sICAM-1 concentration (ng/mL) O D =4 50 n m 0.01 0.1 1 10 0.1 1 10 100 Assay Diluent B Human sICAM-1 concentration (ng/mL) O D =4 50 n m 0.01 0.1 1 10 0.1 1 10 100 Sensitivity: The minimum detectable dose of sICAM-1 is typically less than 150 pg/mL. Recovery: Recovery was determined by spiking various levels of human sICAM-1 into human serum, plasma and cell culture media. Mean recoveries are as follows: Sample Type Average % Recovery Range (%) Serum 97.48 88-106 Plasma 98.67 90-107 Cell culture media 97.37 89-107 Linearity: Sample Type Serum Plasma Cell Culture Media 1:2 Average % of Expected 98 97 96 Range (%) 91-106 89-105 89-106 1:4 Average % of Expected 97 95 96 Range (%) 89-106 88-105 90-107 Reproducibility: Intra-Assay: CV<10 % Inter-Assay: CV<12 %
Assay Precision:	Intra-Assay: CV< 10 % Inter-Assay: CV< 12 %
Restrictions:	For Research Use only

Handling

Handling Advice:	Avoid repeated freeze-thaw cycles.
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

Publications

Product cited in:	Wu, Ding, Han, Arriens, Wei, Han, Pedroza, Jiang, Anolik, Petri, Sanz, Saxena, Mohan: "Antibody-Array-Based Proteomic Screening of Serum Markers in Systemic Lupus Erythematosus: A

Discovery Study." in: **Journal of proteome research**, Vol. 15, Issue 7, pp. 2102-14, (2016) (PubMed).

Tawaramoto, Kaneto, Hashiramoto, Kawasaki, Tatsumi, Shimoda, Kamei, Matsuki, Mune, Kaku et al.: "Azelnidipine, but not amlodipine, reduces urinary albumin excretion and carotid atherosclerosis in subjects with type 2 diabetes: blood pressure control with olmesartan and azelnidipine in Type 2 ..." in: **Diabetology & metabolic syndrome**, Vol. 7, pp. 80, (2015) (PubMed).

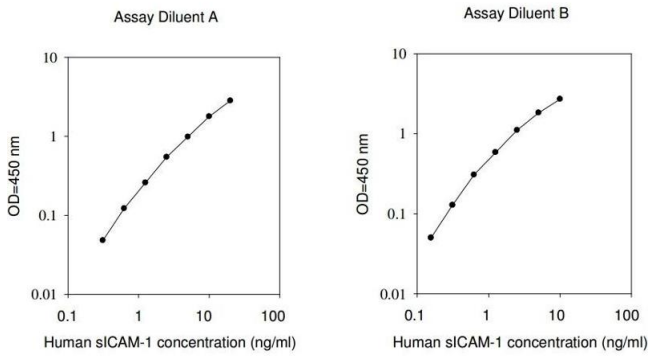
Nirala, Gohil: "Effect of garlic component s-allyl cysteine sulfoxide on glycated human serum albumin induced activation of endothelial cells: an in vitro study." in: **European review for medical and pharmacological sciences**, Vol. 19, Issue 11, pp. 2125-31, (2015) (PubMed).

Zhang, Lin, Jiang, Xu, Luo, Mo, Li, Chen: "Extensive serum biomarker analysis in patients with ST segment elevation myocardial infarction (STEMI)." in: **Cytokine**, Vol. 76, Issue 2, pp. 356-62, (2015) (PubMed).

Liu, Ho, Chen, Woo: "Effect of soy protein and isoflavones on blood pressure and endothelial cytokines: a 6-month randomized controlled trial among postmenopausal women." in: **Journal of hypertension**, Vol. 31, Issue 2, pp. 384-92, (2013) (PubMed).

There are more publications referencing this product on: Product page

Images



ELISA

Image 1.