

Datasheet for ABIN625186

VEGFA ELISA Kit

1 Image

40 Publications



[Go to Product page](#)

Overview

Quantity: 96 tests

Target: VEGFA

Reactivity: Mouse

Method Type: Sandwich ELISA

Detection Range: 2-1000 pg/mL

Minimum Detection Limit: 2 pg/mL

Application: ELISA

Product Details

Purpose: Mouse VEGF-A 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, sTNF α , TARC, TIMP-1, TNF-alpha, Tpo.

Cross-Reactivity (Details): This ELISA kit shows no cross-reactivity with any of the cytokines tested (e.g., 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-1alpha, MIP-2, MIP-3beta, RANTES, SCF, sTNF α , TARC, TIMP-1, TNF-alpha, Tpo).

Product Details

Sensitivity: < 2 pg/mL

Characteristics:

- Strip plates and additional reagents allow for use in multiple experiments
- Quantitative protein detection
- Establishes normal range
- 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: VEGFA

Alternative Name: VEGF-A ([VEGFA Products](#))

Background: Vascular endothelial growth factor A (VEGF-A) (Vascular permeability factor) (VPF)

Gene ID: 22339

UniProt: [Q00731](#)

Pathways: [RTK Signaling](#), [Glycosaminoglycan Metabolic Process](#), [Regulation of Cell Size](#), [Tube Formation](#), [Signaling Events mediated by VEGFR1 and VEGFR2](#), [Platelet-derived growth Factor Receptor Signaling](#), [VEGFR1 Specific Signals](#), [VEGF Signaling](#)

Application Details

Application Notes: Recommended Dilution for serum and plasma samples 3 fold

Application Details

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. 1x Assay Diluent B (Item E) should be used for dilution of culture supernatants. Suggested dilution for normal serum/plasma: 3 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. 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 25 ng/mL standard. Dissolve the powder thoroughly by a gentle mix. Add 40 µL VEGF standard from the vial of Item C, into a tube with 960 µL Assay Diluent A or 1x Assay Diluent B to prepare a 1,000 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 . Mix each tube thoroughly before the next transfer. Assay Diluent A or 1x Assay Diluent B serves as the zero standard (0 pg/mL).
200 µL 200 µL 200 µL 200 µL 200 µL 40 µL standard + 960.0 µL
200µl 1,000 400 160 64 25.6 10.2 4.1 0 pg/mL pg/mL pg/mL pg/mL pg/mL pg/mL pg/mL
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 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) before use. HRP-Streptavidin concentrate should be diluted 160-fold with 1x Assay Diluent B. For example: Briefly spin the vial (Item G) and pipette up and down to mix gently. Add 50 µL of HRP-Streptavidin concentrate into a tube with 8 ml 1x Assay Diluent B to prepare a 160-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 µl) 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 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 Mouse VEGF concentration (pg/mL) 1 10 100 1000 10000
O D = 0.01 0.1 1 10 100
Assay Diluent B Mouse VEGF concentration (pg/mL) 1 10 100 1000
10000
O D = 0.1 1 10

Sensitivity: The minimum detectable dose of VEGF is typically less than 2 pg/mL.

Recovery: Recovery was determined by spiking various levels of mouse VEGF into mouse serum, plasma and cell culture media. Mean recoveries are as follows: Sample Type Average %

Application Details

Recovery Range (%) Serum 87.67 79-102 Plasma 89.32 81-103 Cell culture media 94.29 83-105

Linearity: Sample Type Serum Plasma Cell Culture Media 1:2 Average % of Expected 96 97 95 Range (%) 83-103 84-103 82-102 1:4 Average % of Expected 94 98 97 Range (%) 82-102 84-104 83-103

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: Basu, Bhattacharjee, Ghosh, Samanta, Bhattacharya: "Sensitizing effects of an organovanadium compound during adjuvant therapy with cyclophosphamide in a murine tumor model." in: **Biomedicine & pharmacotherapy**, Vol. 93, pp. 816-829, (2018) ([PubMed](#)).

Huey, Smith, Sulaeman, Breen: "Skeletal myofiber VEGF is necessary for myogenic and contractile adaptations to functional overload of the plantaris in adult mice." in: **Journal of applied physiology (Bethesda, Md. : 1985)**, Vol. 120, Issue 2, pp. 188-95, (2016) ([PubMed](#)).

Kondo, Tsunematsu, Yamada, Arakaki, Saito, Otsuka, Kujiraoka, Ushio, Kurosawa, Kudo, Ishimaru: "Acceleration of tumor growth due to dysfunction in M1 macrophages and enhanced angiogenesis in an animal model of autoimmune disease." in: **Laboratory investigation; a journal of technical methods and pathology**, Vol. 96, Issue 4, pp. 468-80, (2016) ([PubMed](#)).

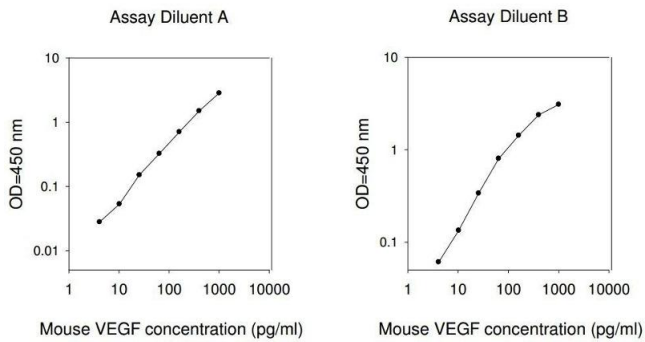
Li, Wang, Huang, Zhang, Yao, Wang, Lv, An, Corrigan, Huang, Ying: "Distinct sustained structural and functional effects of interleukin-33 and interleukin-25 on the airways in a murine asthma

surrogate." in **Immunology**, Vol. 145, Issue 4, pp. 508-18, (2015) ([PubMed](#)).

Pala, Atilgan, Ozkan, Kavak, Ilhan, Akpolat, Sapmaz: "Effect of varying doses of tamoxifen on ovarian histopathology, serum VEGF, and endothelin 1 levels in ovarian hyperstimulation syndrome: an experimental study." in: **Drug design, development and therapy**, Vol. 9, pp. 1761-6, (2015) ([PubMed](#)).

There are more publications referencing this product on: [Product page](#)

Images



ELISA

Image 1.