

Datasheet for ABIN625273 Cathepsin S ELISA Kit

Image



#### Overview

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| Quantity:                | 96 tests           |
|--------------------------|--------------------|
| Target:                  | Cathepsin S (CTSS) |
| Reactivity:              | Human              |
| Method Type:             | Sandwich ELISA     |
| Detection Range:         | 4 pg/mL-1000 pg/mL |
| Minimum Detection Limit: | 4 pg/mL            |
| Application:             | ELISA              |

#### Product Details

| Purpose:           | Human Cathepsin S 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 the following cytokines tested: human Angiogenin          |
|                    | BDNF, BLC, CNTF, ENA- 78, FGF-4, IL-1 alpha, IL-1 beta, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, |
|                    | IL-11, IL-12 p70, IL-12 p40, IL-13, IL-15, I-309, IP-10, FGF-4, FGF-6, FGF- 7, G-CSF, GDNF, GM-         |
|                    | CSF, IFN-gamma, IGFBP-2, IGFBP-3, IGFBP-4, Leptin (OB), MCP-1, MCP-2, MCP-3, MDC, MIF,                  |
|                    | MIG, MIP-1 alpha, MIP-1 beta, MIP-1 delta, PARC, PDGF, RANTES, SCF, SDF-1 alpha, TARC, TGF              |
|                    | beta, TIMP-1, TIMP-2, TNF-alpha, TNF-beta, TPO, VEGF  |
| Sensitivity:       | < 4 pg/mL   |

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### Product Details

| Characteristics:       | <ul> <li>Strip plates and additional reagents allow for use in multiple experiments</li> <li>Quantitative protein detection</li> <li>Establishes normal range</li> <li>The best products for confirmation of antibody array data</li> </ul>  |
|------------------------|--|
| Components:            | <ul> <li>Pre-Coated 96-well Strip Microplate</li> <li>Wash Buffer</li> <li>Stop Solution</li> <li>Assay Diluent(s)</li> <li>Lyophilized Standard</li> <li>Biotinylated Detection Antibody</li> <li>Streptavidin-Conjugated HRP</li> <li>TMB One-Step Substrate</li> </ul>  |
| Material not included: | <ul> <li>Distilled or deionized water</li> <li>Precision pipettes to deliver 2 µL to 1 µL volumes</li> <li>Adjustable 1-25 µL pipettes for reagent preparation</li> <li>100 µL and 1 liter graduated cylinders</li> <li>Tubes to prepare standard and sample dilutions</li> <li>Absorbent paper</li> <li>Microplate reader capable of measuring absorbance at 450nm</li> <li>Log-log graph paper or computer and software for ELISA data analysis</li> </ul> |

| Target:           | Cathepsin S (CTSS)  |
|-------------------|---|
| Alternative Name: | Cathepsin S (CTSS Products)   |
| Background:       | The Human Cathepsin S ELISA (Enzyme-Linked Immunosorbent Assay) kit is an in vitro                |
|                   | enzyme-linked immunosorbent assay for the quantitative measurement of human Cathepsin S           |
|                   | in serum, plasma, cell culture supernatants and urine. This assay employs an antibody specific    |
|                   | for human Cathepsin S coated on a 96-well plate. Standards and samples are pipetted into the      |
|                   | wells and Cathepsin S present in a sample is bound to the wells by the immobilized antibody.      |
|                   | The wells are washed and biotinylated anti-human Cathepsin S 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 Cathepsin S bound. The Stop Solution changes the          |
|                   | color from blue to yellow, and the intensity of the color is measured at 450 nm. Reproducibility: |
|                   | Intra-Assay: CV<10% Inter-Assay: CV<12%.  |
| Gene ID:          | 1520  |
|                   |   |

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# Target Details

| UniProt:  | P25774   |
|-----------|--|
| Pathways: | Activation of Innate immune Response, Toll-Like Receptors Cascades |

# Application Details

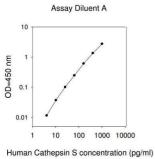
| Application Notes:   | Recommended Dilution for serum and plasma samples50 - 500 fold  |
|----------------------|---|
| Sample Volume:       | 100 µL  |
| Plate:               | Pre-coated  |
| Protocol:            | 1. Prepare all reagents, samples and standards as instructed in the manual.                                 |
|                      | 2. Add 100 $\mu$ L of standard or sample to each well.  |
|                      | 3. Incubate 2.5 h at RT or O/N at 4 °C.   |
|                      | 4. Add 100 $\mu$ L of prepared biotin antibody to each well.  |
|                      | 5. Incubate 1 h at RT.  |
|                      | 6. Add 100 $\mu$ L 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 and urine. Suggested dilution for normal serum/plasma: 50-500 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 before use.                   |
|                      | 4. Preparation of standard: Briefly spin the vial of Item C and then add 400 $\mu L$ Assay Diluent A        |
|                      | (for serum/plasma samples) or 1x Assay Diluent B (for cell culture supernates/urine) into Item              |
|                      | C vial to prepare a 50 ng/mL standard. Dissolve the powder thoroughly by a gentle mix. Add 20               |
|                      | $\mu L$ Cathepsin S standard (50 ng/mL) from the vial of Item C, into a tube with 980 $\mu L$ Assay         |
|                      | Diluent A or 1x Assay Diluent B to prepare a 1,000 pg/mL standard solution. Pipette 300 $\mu L$             |
|                      | Assay Diluent A or 1x Assay Diluent B into each tube. Use the 1,000 pg/mL standard solution to              |
|                      | produce a dilution series . Mix each tube thoroughly before the next transfer. Assay Diluent A o            |
|                      | 1x Assay Diluent B serves as the zero standard (0 pg/mL). 20 $\mu$ L standard + 980 $\mu$ L 200 $\mu$ L 200 |
|                      | μL 200 μL 200 μL 200 μL 200myl 1,000 400 160 64 25.60 10.24 4.10 0 pg/mL pg/mL pg/mL                        |
|                      | pg/mL pg/mL pg/mL pg/mL   |

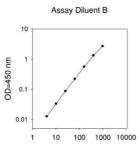
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|                         | 5. If the Wash Concentrate (20x) (Item B) contains visible crystals, warm to room temperature          |
|-------------------------|--|
|                         | and mix gently until dissolved. Dilute 20 ml of Wash Buffer 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 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 300-fold with 1x Assay               |
|                         | Diluent B. For example: Briefly spin the vial (Item G) and pipette up and down to mix gently . Add     |
|                         | 40 µL of HRP-Streptavidin concentrate into a tube with 12 ml 1x Assay Diluent B to prepare a           |
|                         | 300-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                 |
|                         | minutes at room temperature in the dark with gentle shaking.   |
|                         | 9. Add 50 $\mu$ 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.                            |

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|                  | Typical Data: These standard curves are for demonstration only. A standard curve must be run     |
|------------------|--|
|                  | with each assay. Assay Diluent A Human Cathepsin S concentration (pg/mL) 1 10 100 1000           |
|                  | 10000 O D =4 50 n m 0.01 0.1 1 10 Assay Diluent B Human Cathepsin S concentration (pg/mL)        |
|                  | 1 10 100 1000 0 D =4 50 n m 0.01 0.1 1 10  |
|                  | Sensitivity: The minimum detectable dose of Cathepsin S is typically less than 4 pg/mL.          |
|                  | Recovery: Recovery was determined by spiking various levels human Cathepsin S into human         |
|                  | serum, plasma and cell culture media. Mean recoveries are as follows: Sample Type Average $\%$   |
|                  | Recovery Range ( %) Serum 126.3 106-138 Plasma 127.8 115-140 Cell culture media 128.5            |
|                  | 104-139  |
|                  | Linearity: Sample Type Serum Plasma Cell Culture Media 1:2 Average % of Expected 98.71           |
|                  | 97.14 98.81 Range ( %) 87-106 90-104 90-107 1:4 Average % of Expected 97.13 103.0 90.67          |
|                  | Range ( %) 88-106 95-111 85-96   |
|                  | 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   |
|                  |  |





thepsin S concentration (pg/ml) Human Cathepsin S concentration (pg/ml)

# ELISA

Image 1.