Datasheet for ABIN955786

CD44 Standard ELISA Kit

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Overview

Quantity:	96 tests
Target:	CD44 Standard (CD44s)
Reactivity:	Human
Method Type:	Sandwich ELISA
Application:	ELISA

Product Details

Sample Type:	Cell Culture Supernatant, Plasma, Serum, Urine
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	The interference of circulating factors of the immune system was evaluated by spiking various proteins (listed below) at physiologically relevant concentrations into a sCD44std positive serum. There was no detectable cross reactivity.
Sensitivity:	The limit of detection for sCD44std, defined as the analyte concentration resulting in an absorption significantly higher than the absorption of the dilution medium (mean plus two standard deviations), was determined to be 0.02 ng/mL (mean of 6 independent assays).
Characteristics:	The Human sCD44std ELISA detects all circulating CD44 isoforms containing the standard protein sequences (black area).An anti-sCD44std monoclonal coating antibody is adsorbed onto microwells. sCD44std present in the sample or calibrator binds to antibodies adsorbed to the microwells, a HRP-conjugated monoclonal anti-sCD44std antibody is added and binds to sCD44std captured by the first antibody. Coated Microwell - Monoclonal Coating Antibody First Incubation - sCD44std - HRP-Conjugate Following incubation unbound enzyme conjugated anti-

Product Details

	sCD44std is removed during a wash step and substrate solution reactive with HRP is added to the wells. - sCD44std - HRP-Conjugate A colored product is formed in proportion to the amount of sCD44std present in the sample. The reaction is terminated by addition of acid and absorbance is measured at 450 nm. A calibration curve is prepared from six sCD44std Calibrator dilutions that allows determination of sCD44std concentration in samples.
Components:	1 aluminum pouch with a Microwell Plate coated with monoclonal antibody (mouse) to human sCD44std 2 vials (10 µL) HRP-Conjugate anti-sCD44std monoclonal (mouse) antibody 2 vials (0.3 mL) 8 ng/mL sCD44std Calibrator 1 bottle (50 mL) Wash Buffer Concentrate 20X, phosphate-buffered saline (PBS) with 1% Tween 20 1 vial (5 mL) Assay Buffer Concentrate 20X, PBS with 1% Tween 20 and protein stabilizer 1 bottle (60 mL) Sample Diluent 1 vial (15 mL) Substrate Solution 1 vial (12 mL) Stop Solution (1M Phosphoric acid) 1 vial (0.4 mL) Blue Dye 1 vial (0.4 mL) Green Dye 2 adhesive Plate Seals.
Material not included:	5 mL and 10 mL graduated pipettes 5 µL to 1,000 µL adjustable single channel micropipettes with disposable tips 50 µL to 300 µL adjustable multi-channel micropipette with disposable tips Multi-channel micropipette reservoir Beakers, flasks, cylinders

Target Details

Target:	CD44 Standard (CD44s)
Alternative Name:	sCD44std (CD44s Products)
Background:	CD44 (Pgp-1, Ly-24, ECMR III, F10-44-2, H-CAM, HUTCH-I, In(Lu)-related p80, Hermes antigen, hyaluronan receptor) is a polymorphic glycoprotein with apparent molecular weights ranging from 85 kDa to 250 kDa. This cell membrane associated molecule has a cytoplasmic tail (mediates the interaction with the cytoskeleton), a short hydrophobic transmembrane region and an NH2-terminal extracellular (binds to hyaluronate) domain. CD44 isoforms participate in a wide variety of cell-cell or cell-matrix interactions including lymphocyte homing, establishment of B and T cell immune responses, tumor metastases formation and

Target Details

inflammation. Three isoform categories of the CD44 molecule have been identified: 1. an 80-90 kDa isoform, the so-called standard form named CD44std, which is widely distributed on several hematopoietic and non-hematopoietic cells including all subsets of leukocytes, monocytes, erythrocytes, many types of epithelium, mesenchymal elements like fibroblasts, smooth muscle cells and glial cells of the central nervous system, 2. a medium size category of 110-160 kDa which is weakly expressed on epithelial cells and highly expressed in some carcinomas and 3. a category which includes very large isoforms of 250 kDa covalently modified by the addition of chondroitin sulfate. These larger isoforms of CD44 arise by alternative splicing of one or more variant exons (v2-v10) into the extracellular part of the 90 kDa constant form molecule. Compared to the standard CD44, all larger isoforms are expressed in a much more restricted fashion, only in a few normal tissues or on the surface of certain tumor cells. Some splice variants of CD44 play important and distinct roles in tumor metastasis.

Application Details

Plate:	Pre-coated
Reagent Preparation:	Buffer Concentrates should be brought to room temperature and should be diluted before starting the test procedure. If crystals have formed in the Buffer Concentrates, warm them gently until they have completely dissolved. Wash Buffer: Pour entire contents (50 mL) of the Wash Buffer Concentrate (20X) into a clean 1,000 mL graduated cylinder. Bring final volume to 1,000 mL with glass-distilled or de-ionized water. Mix gently to avoid foaming. The pH of the final solution should adjust to 7.4. Transfer to a clean wash bottle and store at 2-25°C. Please note that Wash Buffer is stable for 30 days. Assay Buffer: Pour the entire contents (5 mL) of the Assay Buffer Concentrate (20X) into a clean 100 mL graduated cylinder. Bring to final volume of 100 mL with distilled water. Mix gently to avoid foaming. Store at 4 °C. Please note that the Assay Buffer (1X) is stable for 30 days. Preparation of HRP-Conjugate: Dilute the HRP-Conjugate 1:50 just prior to use by adding 490 µL of (1X) Assay Buffer to the tube containing the HRP- Conjugate concentrate. Mix the contents of the tube well. Make a further 1:40 dilution with Assay Buffer (1X) in a clean plastic tube or reagent reservoir. Please note that the HRP-Conjugate should be used within 30 minutes after dilution.

Application Details

	Addition of color-giving reagents: Blue Dye, Green Dye (OPTIONAL STEP): In order to help our customers to avoid any mistakes in pipetting, now offers a new tool that helps to monitor the addition of even very small volumes of a solution to the reaction well by giving distinctive colors to each step of the ELISA procedure. This procedure is optional, does not in any way interfere with the test results, and is designed to help the customer with the performance of the test, but can also be omitted.
Sample Preparation:	Cell culture supernatants, human serum, EDTA, citrate, or heparinized plasma, amniotic fluid, urine, or other body fluids are suitable for use in the assay. Remove the serum or plasma from the clot or red cells, respectively, as soon as possible after clotting and separation. Samples containing a visible precipitate must be clarified prior to use in the assay. Do not use grossly hemolyzed or lipemic specimens. Samples should be aliquoted and must be stored frozen at -20°C to avoid loss of bioactive sCD44std. If samples are to be run within 24 hours, they may be stored at 4°C. Avoid repeated freeze-thaw cycles. Prior to assay, frozen sera, plasma and urine samples should be brought to room temperature (RT) slowly and mixed gently.
Restrictions:	For Research Use only

Handling

Handling Advice:	Since exact conditions may vary from assay to assay, a calibration curve must be established for every run. Bacterial or fungal contamination of either screen samples or reagents or cross-contamination between reagents may cause erroneous results.
Storage:	4 °C
Storage Comment:	Store kit reagents at 4°C. Immediately after use reagents should be returned to cold storage (4°C). Expiration date of the kit and reagents is stated on the labels. The expiration date of the kit components can only be guaranteed if the components are stored properly, and if, in case of repeated use of one component, the reagent is not contaminated by the first handling.
Expiry Date:	The expiry date is stated on the label.