Datasheet for ABIN956343

ABCD1 ELISA Kit

Go to Product page

Overview

Quantity:	96 tests
Target:	ABCD1
Reactivity:	Rat
Method Type:	Competition ELISA
Detection Range:	24.69-2000 pg/mL
Minimum Detection Limit:	24.69 pg/mL
Application:	ELISA

Product Details

Purpose:	The kit is a competitive inhibition enzyme immunoassay technique for the in vitro quantitative measurement of rat ALD in serum, plasma and other biological fluids.
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This assay has high sensitivity and excellent specificity for detection of rat ALD. No significant cross- reactivity or interference between rat ALD and analogues was observed. Note: Limited by current skills and knowledge, it is impossible for us - complete the cross-reactivity detection between rat ALD and all the analogues, therefore, cross reaction may still exist.
Sensitivity:	The minimum detectable dose of rat ALD is typically less than 9.35 pg/mL. The Sensitivity of this assay, or Lower Limit of Detection (LLD) was defined as the lowest protein concentration that could be differentiated from zero. It was determined the mean O.D. Value of 20 replicates of the zero calibrator plus three standard deviations.

Product Details

Characteristics:	This assay employs the competitive inhibition enzyme immunoassay technique. A monoclonal antibody specific for rat ALD has been pre-coated onto a microplate. A competitive inhibition reaction is launched between biotin labeled rat ALD and unlabeled rat ALD (Calibrators or samples) with the pre-coated antibody specific for rat ALD. After incubation the unbound conjugate is washed off. Next, avidin conjugated to Horseradish Peroxidase (HRP) is added to each microplate well and incubated. The amount of bound HRP conjugate is reverse proportional to the concentration of ALD in the sample. After addition of the substrate solution, the intensity of color developed is reverse proportional to the concentration of ALD in the sample.
Components:	Pre-coated, ready to use 96-well strip plate (1x) Calibrator (lyophilized) (2x) Calibrator Diluent (1 x 20 mL) Detection Reagent A (1 x 120 µL) Detection Reagent B (1 x 120 µL) Assay Diluent A (2X concentrate) (1 x 6 mL) Assay Diluent B (2X concentrate) (1 x 6 mL) TMB Substrate (1 x 9 mL) Stop Solution (1 x 6 mL) Wash Buffer (30X concentrate) (1 x 20 mL) Plate sealer for 96 wells (4x).
Material not included:	1. Microplate reader with 450 +/- 10 nm filter. 2. Precision single and multi-channel pipettes and disposable tips. 3. Eppendorf Tubes for diluting samples. 4. De-ionized or distilled water. 5. Absorbent paper for blotting the microtiter plate. 6. Container for Wash Solution.

Target Details

Target:	ABCD1
Alternative Name:	ALD (ABCD1 Products)
Pathways:	Monocarboxylic Acid Catabolic Process

Application Details

Comment:	The calibration curve concentrations used for the ELISA's were 2, 000 pg/mL, 666.67 pg/mL, 222.22 pg/mL, 74.07pg/mL, 24.69 pg/mL.
Plate:	Pre-coated
Assay Procedure:	1. Determine wells for diluted calibrator, blank and sample. Prepare 5 wells for calibrator, 1 well for blank. Add 50 µL each of dilutions of calibrator (read Reagent Preparation), blank and samples into the appropriate wells, respectively. And then add 50 µL of Detection Reagent A to each well immediately. Shake the plate gently (using a microplate shaker is recommended). Cover with a Plate sealer. Incubate for 1 hour at 37° C. Detection Reagent A may appear cloudy. Warm to room temperature and mix gently until solution appears uniform. 2. Aspirate the solution and wash with 350 µL of 1X Wash Solution to each well using a squirt bottle, multi-channel pipette, manifold dispenser or autowasher, and let it sit for 1~2 minutes. Remove the remaining liquid from all wells completely by snapping the plate onto absorbent paper. Repeat 3 times. After the last wash, remove any remaining Wash Buffer by aspirating or decanting. Invert the plate and blot it against absorbent paper. 3. Add 100 µL of Detection Reagent B working solution to each well. Incubate for 30 minutes at 37° C after covering it with the Plate sealer. 4. Repeat the aspiration/wash process for five times as conducted in step 2. 5. Add 90 µL of Substrate Solution to each well. Cover with a new Plate sealer. Incubate for 10 15 minutes at 37° C. Protect from light. The liquid will turn blue by the addition of Substrate Solution. 6. Add 50 µL of Stop Solution to each well. The liquid will turn yellow by the addition of Stop solution. Mix the liquid by tapping the side of the plate. If color change does not appear uniform, gently tap the plate to ensure thorough mixing. 7. Remove any drop of water and fingerprint on the bottom of the plate and confirm there is no bubble on the surface of the liquid. Then, run the microplate reader and conduct measurement at 450 nm immediately. Note: 1. Assay preparation: Keep appropriate numbers of strips for 1 experiment and remove extra strips from microtiter plate. Removed strips should be resealed and stored at -20°C until the kit expiration date. 2. Samples or reagents addition: Please use the freshly prepared Calibrator. Please carefully add samples to wells and mix gently to avoid foaming. Do not touch the well wall as possible. For each step in the procedure, total dispensing time for addition of reagents or samples to the assay plate should not exceed 10 minutes. This will ensure equal elapsed time for each

Application Details

	pipetting step, without interruption. Duplication of all calibrators and specimens, although not required, is recommended. To avoid cross-contamination, change pipette tips between additions of each calibrator level, between sample additions, and between reagent additions. Also, use separate reservoirs for each reagent. 3. Incubation: To ensure accurate results, proper adhesion of plate sealers during incubation steps is necessary. Do not allow wells to sit uncovered for extended periods between incubation steps. Once reagents have been added to the well strips, DO NOT let the strips DRY at any time during the assay. Incubation time and temperature must be observed. 4. Washing: The wash procedure is critical. Complete removal of liquid at each step is essential to good performance. After the last wash, remove any remaining Wash Solution by aspirating or decanting and remove any drop of water and fingerprint on the bottom of the plate. Insufficient washing will result in poor precision and falsely elevated absorbance reading. 5. Controlling of reaction time: Observe the change of color after adding TMB Substrate (e.g. observation once every 10 minutes), if the color is too deep, add Stop Solution in advance to avoid excessively strong reaction which will result in inaccurate absorbance reading. 6. TMB Substrate is easily contaminated. Please protect it from light. 7. The environment humidity which is less than 60% might have some effects on the final performance, therefore, a humidifier is recommended to be used at that condition.
Calculation of Results:	This assay employs the competitive inhibition enzyme immunoassay technique, so there is an inverse correlation between ALD concentration in the sample and the assay signal intensity. Average the duplicate readings for each calibrator, control, and samples. Create a calibration curve on log-log or semi-log graph paper, with the log of ALD concentration on the y-axis and absorbance on the x- axis. Draw the best fit straight line through the calibrator points and it can be determined by regression analysis. Using some plot software is also recommended. If samples have been diluted, the concentration read from the calibration curve must be multiplied by the dilution factor.
Restrictions:	For Research Use only

Handling

Storage:	-20 °C
Storage Comment:	All the reagents should be kept according to the labels on vials. The Calibrator, Detection Reagent A, Detection Reagent B and the 96-well strip plate should be stored at -20° C upon being received. The unused strips should be kept in a sealed bag with the desiccant provided to minimize exposure to damp air. Opened test kits will remain stable until the expiration date

Handling

	shown, provided it is stored as prescribed above.
Expiry Date:	The expiry date is stated on the label.