Type II Aldolase (ALD2) ELISA Assay Kit

Cat. No. KITG-014 **Lot. No.** (See product label)

SPECIFICATION

Product Overview	This kit is used for the determination of Type II Aldolase (ALD2) activity in microbial samples.
Description	This kit uses a double-antibody sandwich method to determine Type II Aldolase (ALD 2) levels in specimens. Purified Type II Aldolase (ALD2) antibody is coated on micropl ates to form a solid-phase antibody. Type II Aldolase (ALD2) is added sequentially to the monoclonal antibody-coated wells, followed by binding with HRP-labeled Type II Aldolase (ALD2) antibody to form an antibody-antigen-enzyme-labeled antibody comp lex. After thorough washing, the substrate TMB is added for color development. TMB i s converted to blue under the catalysis of HRP enzyme and to the final yellow under t he action of acid. The color intensity is positively correlated with the Type II Aldolase (ALD2) in the sample. The absorbance (OD value) is measured at 450 nm wavelength using a microplate reader, and the Type II Aldolase (ALD2) activity concentration in th e sample is calculated through the standard curve.
Form	ELISA Assay Kit
Applications	Determination of Type II Aldolase (ALD2) activity in microbial samples
Notes	Kit Components: Component 1: 30× Concentrated Wash Buffer, 20 mL in 1 vial Comp onent 2: Enzyme Reagent, 6 mL in 1 vial Component 3: Antibody-coated Microplate, 12 wells by 8 strips Component 4: Sample Diluent, 6 mL in 1 vial Component 5: Chro mogen A Solution, 6 mL in 1 vial Component 6: Chromogen B Solution, 6 mL in 1 vial Component 7: Stop Solution, 6 mL in 1 vial Component 8: Standard at 16 IU/L concen

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tration, 0.5 mL in 1 vial Component 9: Standard Diluent, 1.5 mL in 1 vial Component 10: Instruction Manual, 1 copy Component 11: Plate Sealer, 2 sheets Component 12: Sealing Bag, 1 piece Samples Requirements: 1. Specimens should be extracted as soon as possible after collection. Follow relevant literature for extraction procedures. Experiments should be conducted promptly after extraction. If testing cannot be performed immediately, specimens may be stored at minus 20 degrees Celsius, but repeated freeze-thaw cycles should be avoided. 2. Samples containing NaN3 cannot be detected, as NaN3 inhibits horseradish peroxidase (HRP) activity. Assay Protocol: 1. Standard Dilution: This kit provides one vial of stock standard. Users may perform dilutions in small test tubes according to the following procedure: Standard 5 at 8 IU/L: Mix 150 microliters of stock standard with 150 microliters of standard diluent. Standard 4 at 4 IU/L: Mix 150 microliters of Standard 5 with 150 microliters of standard diluent. Standard 3 at 2 IU/L: Mix 150 microliters of Standard 4 with 150 microliters of standard diluent. Standard 2 at 1 IU/L: Mix 150 microliters of Standard 3 with 150 microliters of standard diluent. Standard 1 at 0.5 IU/L: Mix 150 microliters of Standard 2 with 150 microliters of standard diluent. 2. Sample Loading: Set up blank wells (blank control wells receive no sample or enzyme reagent; all other steps are the same), standard wells, and sample wells to be tested. Add 50 microliters of standard accurately to the standard wells on the antibody-coated plate. For sample wells, first add 40 microliters of sample diluent, then add 10 microliters of sample to be tested (final sample dilution is 5-fold). Add samples to the bottom of the microplate wells, avoiding contact with the well walls, and mix gently by swirling. 3. Incubation: Seal the plate with plate sealer and incubate at 37 degrees Celsius for 30 minutes. 4. Buffer Preparation: Dilute the 30× concentrated wash buffer 30-fold with distilled water for use. 5. Washing: Carefully remove the plate sealer, discard the liquid, and shake dry. Fill each well with wash buffer, let stand for 30 seconds, then discard. Repeat this process 5 times, then pat dry. 6. Enzyme Addition: Add 50 microliters of enzyme reagent to each well, except the blank wells. 7. Incubation: Same as step 3. 8. Washing: Same as step 5. 9. Color Development: First add 50 microliters of Chromogen A to each well, then add 50 microliters of Chromogen B. Mix gently by shaking, and incubate at 37 degrees Celsius protected from li

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ght for 10 minutes. 10. Termination: Add 50 microliters of stop solution to each well. 1 1. Measurement: Zero with the blank well, and measure the absorbance (OD value) o f each well sequentially at 450 nanometers wavelength. Measurement should be perf ormed within 15 minutes after adding the stop solution. Operation Procedure Summar y: Step 1: Prepare reagents, samples, and standards. Step 2: Add prepared samples and standards, react at 37 degrees Celsius for 30 minutes. Step 3: Wash plate 5 time s, add enzyme reagent, react at 37 degrees Celsius for 30 minutes. Step 4: Wash plat e 5 times, add Chromogen A and B, develop color at 37 degrees Celsius for 10 minut es. Step 5: Add stop solution. Step 6: Read OD value within 15 minutes. Step 7: Calc ulate. Calculation: Plot the standard curve with standard concentration as the x-axis a nd OD value as the y-axis on graph paper. Determine the corresponding concentratio n from the standard curve based on the sample OD value, then multiply by the dilutio n factor. Alternatively, calculate the linear regression equation of the standard curve u sing standard concentration and OD values, substitute the sample OD value into the equation to calculate sample concentration, then multiply by the dilution factor to obtai n the actual sample concentration. Precautions: 1. The kit should be equilibrated at ro om temperature for 15 to 30 minutes after removal from refrigerated storage. After op ening, unused plate strips should be stored in the sealing bag. 2. Concentrated wash buffer may crystallize; warming in a water bath during dilution can help dissolve crysta ls, which does not affect results. 3. Use pipettes for all loading steps and calibrate the m regularly to avoid experimental errors. Loading time for each step should preferably be controlled within 5 minutes; for large numbers of specimens, a multi-channel pipett e is recommended. 4. Perform a standard curve with each assay, preferably in duplic ate. If the specimen contains excessively high levels of the analyte (sample OD value greater than the first standard well OD value), dilute the sample with sample diluent b y a certain factor (n-fold) before measurement, and multiply by the total dilution factor (times n times 5) in the calculation. 5. Plate sealers are for single use only to avoid cr oss-contamination. 6. Keep substrates protected from light. 7. Strictly follow the instru ctions; results must be determined by microplate reader readings. 8. All samples, was h buffers, and waste materials should be treated as infectious materials. 9. Compone

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	nts from different kit lots must not be mixed.
Storage	2 to 8 degrees Celsius. Shelf life: 6 months.
Usage	For Research Use Only

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