Human HBsAg ADR ELISA Kit

Cat. No. KITG-102 **Lot. No.** (See product label)

SPECIFICATION

Product Overview	The Human HBsAg ADR ELISA Kit is intended for the quantitative determination of human hepatitis B surface antigen ADR subtype (HBsAg ADR) in human serum, plasma, and related samples. The kit contains a 96-well microplate and has a stated detection range of 0.6 IU/L to 26 IU/L.
Species	Human
Description	Assay Principle This kit uses a double-antibody sandwich enzyme-linked immunosorb ent assay to determine the level of human hepatitis B surface antigen ADR subtype (HBsAg ADR) in samples. Purified antibody is coated onto the microplate to form a soli d-phase antibody. Human HBsAg ADR in the sample is added to the antibody-coated wells and then binds to an HRP-labeled antibody, forming an antibody-antigen-enzym e-labeled antibody complex. After thorough washing, TMB substrate is added for colo r development. TMB is converted to blue under HRP catalysis and then to yellow after addition of the acidic stop solution. The color intensity is positively correlated with the amount of HBsAg ADR in the sample. Absorbance is measured at 450 nm, and the H BsAg ADR concentration is calculated from the standard curve.
Form	Kit
Applications	Quantitative measurement of human hepatitis B surface antigen ADR subtype (HBsAg ADR) in human serum, plasma, and related samples.
Notes	Kit Components 30× Concentrated Wash Solution: 20 mL × 1 bottle. Enzyme

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	Conjugate Reagent: 6 mL × 1 bottle. Antibody-Coated Microplate: 12 wells × 8 strips. Sample Diluent: 6 mL × 1 bottle. Chromogen Solution A: 6 mL × 1 bottle. Chromogen Solution B: 6 mL × 1 bottle. Stop Solution: 6 mL × 1 bottle. Standard, 48 IU/L: 0.5 mL × 1 bottle. Standard Diluent: 1.5 mL × 1 bottle. Instruction Manual: 1 copy. Plate Sealing Film: 2 sheets. Sealing Bag: 1. Sample Requirements 1. Process samples as soon as possible after collection according to the applicable extraction method and perform the assay promptly. If immediate testing is not possible, store samples at -20°C and avoid repeated freeze-thaw cycles. 2. Do not test samples containing NaN_3 because sodium azide inhibits horseradish peroxidase activity. Precautions 1. After removal from refrigerated storage, equilibrate the kit at room temperature for 15-30 minutes before use. If the coated plate is not completely used after opening, place the remaining strips in the sealing bag for storage. 2. Crystals may form in the concentrated wash solution. Warm in a water bath during dilution to aid dissolution; this does not affect the washing result. 3. Use calibrated pipettes for all liquid additions. To reduce assay error, complete one round of sample addition within 5 minutes whenever possible. Use a multichannel pipette when many samples are tested. 4. Generate a standard curve for every assay, preferably using duplicate wells. If the sample OD exceeds that of the highest standard well, dilute the sample by n-fold with Sample Diluent and repeat the assay. For the final calculation, multiply by the total dilution factor, n × 5. 5. Plate sealing film is for single use only to prevent cross-contamination. 6. Protect the substrate from light. 7. Follow the instructions strictly; interpretation of assay results must be based on microplate reader measurements. 8. Treat all samples, wash solution, and waste as potentially infectious materials. 9. Do not mix components from different kit lots.
Storage	Store the kit at 2-8°C. The stated shelf life is 6 months.
Reaction conditions	Standard Preparation The kit provides one vial of the original 48 IU/L standard.

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	Prepare the standards in small tubes as follows: 24 IU/L, Standard No. 5: add 150 µL of the original standard to 150 µL of Standard Diluent. 12 IU/L, Standard No. 4: add 150 µL of Standard No. 5 to 150 µL of Standard Diluent. 6 IU/L, Standard No. 3: add 150 µL of Standard No. 4 to 150 µL of Standard Diluent. 3 IU/L, Standard No. 2: add 150 µL of Standard No. 3 to 150 µL of Standard Diluent. 1.5 IU/L, Standard No. 1: add 150 µL of Standard No. 2 to 150 µL of Standard Diluent. Assay Procedure 1. Set blank wells, standard wells, and sample wells. The blank wells receive neither sample nor Enzyme Conjugate Reagent; all other operations are identical. Add 50 µL of the appropriate standard to each standard well. For each sample well, first add 40 µL of Sample Diluent and then 10 µL of sample, giving a final five-fold sample dilution. Add liquid to the bottom of each well without touching the well wall and gently mix. 2. Seal the plate and incubate at 37°C for 30 minutes. 3. Dilute the 30× Concentrated Wash Solution 30-fold with distilled water before use. 4. Carefully remove the sealing film, discard the liquid, and blot dry. Fill each well with wash solution, allow it to stand for 30 seconds, discard the wash solution, and repeat for a total of five washes. Blot the plate dry. 5. Add 50 µL of Enzyme Conjugate Reagent to each well except the blank well. 6. Seal the plate and incubate at 37°C for 30 minutes. 7. Wash the plate five times as described above. 8. Add 50 µL of Chromogen Solution A and then 50 µL of Chromogen Solution B to each well. Gently mix and incubate at 37°C protected from light for 10 minutes. 9. Add 50 µL of Stop Solution to each well. The blue color changes immediately to yellow. 10. Zero the instrument with the blank well and measure the absorbance of each well at 450 nm. Complete the measurement within 15 minutes after adding the Stop Solution.
Unit definition	Result Calculation Plot standard concentration on the horizontal axis and OD value on the vertical axis to generate the standard curve. Determine the corresponding concentration from the sample OD value and multiply by the dilution factor. Alternatively, calculate a linear regression equation from the standard concentrations and OD values, substitute the sample OD value into the equation, and multiply the

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	calculated concentration by the dilution factor. The result is the actual sample concentration. Because the routine sample preparation gives a five-fold dilution, samples diluted by an additional n-fold must be multiplied by n × 5.
Usage	For Research Use Only.

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