

Human Anti-HBV-IgM ELISA Kit

Order details

CatNo: AHY-25

Size: 96T/48T

Product description

Hepatitis B virus (HBV) is an enveloped, double-stranded DNA virus belonging to the Hepadnaviridae family and is recognized as the major cause of blood transmitted hepatitis together with hepatitis C virus (HCV). Infection with HBV induces a spectrum of clinical manifestations ranging from mild, inapparent disease to fulminant hepatitis, severe chronic liver diseases, which in some cases can lead to cirrhosis and carcinoma of the liver. This ELISA kit is an enzyme-linked immunosorbent assay (ELISA) for qualitative detection of antibodies to hepatitis B virus core antigen (anti-HBc) in human serum or plasma. It is intended for use in clinical laboratories for diagnosis and management of patients related to infection with hepatitis B virus.

Classification of a hepatitis B infection requires the identification of a number of serological markers expressed during three phases (incubation, acute and convalescent) of the infection. Now several diagnostic tests are used for screening, clinical diagnosis and management of the disease. Hepatitis B "core" antigen (HBcAg) is a major component of the viral structure. HBcAg is composed of a single polypeptide of about 17 kD that is released upon disaggregation of the core particles; the antigen contains at least one immunological determinant. Antibodies to HBcAg (anti-HBc total antibody, and IgM) appear shortly after the appearance of HBsAg and persist for life both in persons who have recovered from a hepatitis B infection and in those who develop HBsAg-carrier status but in rare cases, a HBV infection can also

run its course without the appearance of immunologically detectable anti-HBc (usually in immunosuppressed patients).

Product description

Microplate	1 plate(12x8/8x12 well strips per plate) each well contains purified HBcAg
Negative Control	1ml,1 vial
Positive Control	1ml,1 vial
HRP-Conjugate	6.5ml,1 vial
Sample Diluent	7ml,1 vial
Substrate Solution A	7ml, 1 vial
Substrate Solution B	7ml, 1 vial
Stop Solution	7ml, 1 vial
Wash Buffer (20×)	30ml, 1 vial
Microtiterplate sealers	1 sheet
Plastic Sealable Bag	1 unit

Storage method: it can be stored at 2-8 °C for more than 6 months. Do not freeze it.

Steps

1. Check the Wash buffer concentrate for the presence of salt crystals. If crystals have formed in the solution, resolubilize by warming at 37°C until crystals dissolve.
2. Set the strips needed in strip-holder and number sufficient number of wells including three Negative control (e.g. B1, C1, D1), two Positive control (e.g. E1, F1) and one Blank (e.g. A1, neither samples nor HRP-Conjugate should be added into the Blank well). If the results will be determined by using dual wavelength plate reader, the requirement for use of Blank well could be omitted. Use only number of strips required for the test.
3. Add Positive and Negative controls into their respective wells, add Sample Diluent into other reaction wells, then add sample, mix well.

Note: Use a separate disposable pipette tip for each specimen, Negative and Positive Control as to avoid cross-contamination.

4. Cover the plate with the plate cover and incubate at 37°C.
5. After the end of the incubation, remove and discard the plate cover. Wash each well 5 times with diluted Washing buffer. Each time allow the microwells to soak. After the final washing cycle, turn down the plate onto blotting paper or clean towel and tap it to remove any remainders.
6. Add Chromogen A and of Chromogen B solutions into each well including the Blank. Incubate the plate at 37°C avoiding light. The enzymatic reaction between the Chromogen solutions and the HRP-Conjugate produces blue color in Negative control and anti-HBc negative sample wells.
7. Using a multichannel pipette or manually, add Stop Solution into each well and mix gently. Intensive yellow color develops in Negative control and anti-HBc negative sample wells.
8. Calibrate the plate reader with the Blank well and read the absorbance at 450 nm. If a dual filter instrument is used, set the reference wavelength at 630 nm. Calculate the Cutoff value and evaluate the results. (Note: read the absorbance within 5 minutes after stopping the reaction.)

Note

1. The kit should be equilibrated to room temperature (20-23°C) before opening any vials and starting the assay. It is highly recommended that the solutions be used as soon as possible after rehydration.
2. When mixing or reconstituting protein solutions, always avoid foaming.
3. Do not mix or substitute reagents with those from other lots or sources.
4. To avoid cross-contamination, change pipette tips between additions of each standard level, between sample additions, and between reagent additions.



5. Crystals could appear in the 20X wash solution due to high salt concentration in the stock solutions. Crystals are readily dissolved at room temperature or at 37°C before dilution of the buffer solutions.
6. Keep TMB Substrate protected from light.
7. The Stop Solution provided with this kit is an acid solution. Wear eye, hand, face, and clothing protection when using this material.