

HISCL™Anti-HBs Assay Kit

REF AV-918-225

Identification of the IVD reagent HISCL™ Anti-HBs Assay Kit

Intended use

For In Vitro Diagnostic Use
Measurement of anti-HBs antibody (HBsAb) in serum or plasma

Development process and characteristics

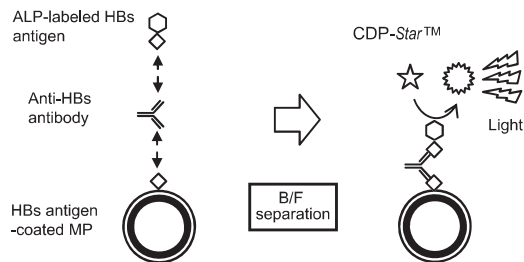
An anti-HBs antibody test is commonly performed for diagnosis of HBV infection.
This kit measures anti-HBs antibody based on the chemiluminescence enzyme immunoassay method with CDP-*Star*™ chemiluminescent substrate.
This kit is exclusively designed for Sysmex automated immunoassay system.

Principles of the examination method

This kit measures anti-HBs antibody based on the 1-step sandwich chemiluminescent enzyme immunoassay.

1. ALP (alkaline phosphatase)-labeled recombinant HBs antigen in R1 reagent and recombinant HBs antigen-coated MP (magnetic particles) in R2 reagent specifically react with anti-HBs antibody in sample.
2. After B/F separation, ALP on MP breaks down CDP-*Star*™ substrate in R5 reagent to an excited intermediate, which produces luminescent signal.

Since the light production increases in proportion to anti-HBs antibody concentration, sample anti-HBs antibody concentrations can be obtained with a calibration curve prepared with calibrators (HISCL Anti-HBs Calibrator).



Components

This kit consists of the following reagents. 3 – 6 are products which are sold separately.

1. R1 reagent: contains ALP-labeled recombinant HBs antigen. 0.43U/mL
2. R2 reagent: contains magnetic particles coated with recombinant HBs antigen. 5 mg/mL
3. HISCL Substrate Reagent Set
 - (1) R4 reagent
 - (2) R5 reagent: contains CDP-*Star*™: Disodium 2-chloro-5-(4-methoxyspiro{1,2-dioxetane-3,2'-(5'-chloro)-tricyclo[3.3.1.1^{3,7}]decan}-4-yl)-1-phenyl phosphate 0.48 mM
4. HISCL Washing solution
5. HISCL Anti-HBs Calibrator
 - (1) HISCL HBsAb C0
 - (2) HISCL HBsAb C1
 - (3) HISCL HBsAb C2
 - (4) HISCL HBsAb C3
6. HISCL Diluent






Warnings and precautions

1. Use the kit according to the method stipulated in the package insert. The reliability of results cannot be guaranteed if the kit is used with a method or for a purpose other than those stipulated.
2. Handle each reagent carefully without generating air bubbles, which may produce incorrect analysis results. If bubbles appear, wait until they disappear.
3. Do not combine reagents from different kits. Do not pool reagents even if the Lot Nos. of kits are the same. Use reagents prior to the expiry date. The reliability of results cannot be guaranteed if reagents are used past their expiration date.
4. Avoid contact of R5 reagent with the skin and eyes, since it is an alkaline solution with pH9.6.
5. All Calibrator bottles should be quickly closed after dispensing drops of Calibrator solution, and then stored at 2-8 °C. If bottles are left open, Calibrators may become concentrated by evaporation, resulting in incorrect calibration.
6. When they are out of the analyzer reagent holder, store R1-R2 reagents at 2-8 °C. Stir R2 reagent according to [Examination procedure] just before you return it to the analyzer. Do not use reagents once they have frozen, since they may exhibit deterioration.
7. The calibration is valid for 30 days. However, even within this period, calibrate again in the following circumstances:
 - When new R1-R2 reagents with another Lot No. are used.
 - When quality assurance results are abnormal.
 - After specified maintenance and/or repair of the analyzer (see analyzer instruction manual).
8. R1 reagent, R2 reagent, R4 reagent, Calibrators and Diluent contain sodium azide. Since sodium azide reacts with lead tubing and copper tubing to generate metal azides which can explode, use a large quantity of water when disposing of it. In case of contact with the eyes, mouth, or hands, carry out emergency treatment such as washing with a large quantity of water. If necessary, consult a physician.
9. HISCL HBsAb C1-C3 contain anti-HBs antigen polyclonal antibody (rabbit), they do not contain human-derived materials.
10. Handle samples carefully. They sometimes contain HBV, HCV, HIV, etc.
11. Do not use the reagent bottles, etc. for other purposes.
12. Use only the reagents (R1 reagents, R2 reagents, R4 reagents, R5 reagents, Calibrators, and Washing solution) specified in this package insert.
13. Be certain to assemble the reagent containers according to [Examination procedure]. Incorrectly assembled containers may result in device errors or cause evaporation of reagents.
14. Install R4 reagent and R5 reagent carefully to prevent contamination by alkaline phosphatase in saliva or on skin. To prevent absorption of excess CO₂, do not remove R5 reagent from the instrument until its bottle is empty and requires replacement.

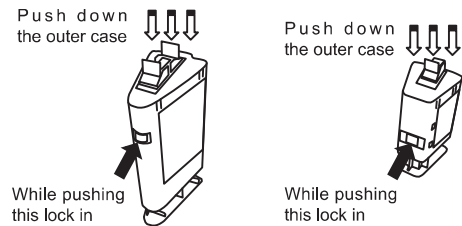
Examination procedure

1. Preparation for measurement
 - (1) Gently mix R2 reagent thoroughly by circling the container. Look and confirm that the magnetic particles have mixed uniformly.



REF	Catalogue number		Use by
IVD	In vitro diagnostic medical device	LOT	Batch code
	Manufacturer		Sufficient for
	Consult instructions for use		
	Temperature limitation		

- (2) First of all push down the outer cases of reagent containers firmly to tear aluminum seals on the inner bottles.



- (3) Set the containers at the indicated position in the analyzer.
- (4) As a rule, dispense 200 µL of sample to reduce possible effects of evaporation. Refer to the analyzer instruction manual for the minimum volume.
2. Standard assay method *
- (1) Dispense 20 µL of sample and 50 µL of R1 reagent into a reaction cuvette, and then incubate for 2 minutes at 42°C.
- (2) Dispense 30 µL of R2 reagent into the cuvette, incubate for 6 minutes at 42 °C, and then carry out magnetic separation (contact the magnet with the cuvette, and aspirate liquid).
- (3) Dispense 100-700 µL of Washing solution, and then carry out magnetic separation. Repeat this procedure 3 times.
- (4) Dispense 50µL of R4 reagent and mix, dispense 100 µL of R5 reagent and mix, incubate for 5 minutes at 42 ° C, and then measure light intensity.
3. Prepare a calibration curve
- (1) Gently stir each of the Calibrators (HISCL HBsAb C0-C3) without generating bubbles. Position them according to the analyzer instruction manual.
- (2) Carry out procedures according to the "standard assay method", and then measure light intensity.
- (3) Plot the intensity of the Calibrators on the ordinate and the Calibrator concentrations on the abscissa, and then prepare a calibration curve.*
4. Sample measurement
- (1) Position a sample according to the analyzer instruction manual.
- (2) Carry out procedures according to the "standard assay method", and then measure light intensity.*
- (3) Fit the intensity on the calibration curve to obtain the anti-HBs antibody concentration in the sample. *

* The analyzer automatically carries out these procedures.

Storage and shelf life after first opening

Store at 2-8 °C. Do not freeze.
The shelf life is 30 days after opening.

Control procedure

Analyze control materials as samples according to [Examination procedure].

Biological reference intervals

Normal sample is negative in this test.

Interpretation of results

The sample is judged positive when the test result is above 5.0 mIU/mL and negative when test result is below 5.0 mIU/mL.

- [Note 1] Do not diagnose hepatitis B virus infection based only on test results obtained with this product. Comprehensive judgment is required, with reference to other test results including determination of HBc antibody, etc., and clinical course.
- [Note 2] Certain samples of HBV vaccine are judged negative in this test when antibody production is low.
- [Note 3] Non-specific reactions can occur in immunoassays. Such reactions are believed to be caused by autoantibodies, insoluble matter (especially fibrin), natural antibodies, etc.
- [Note 4] Assess samples with findings above 1000 mIU/mL as positive. Use HISCL Diluent in case of dilution test. HISCL Diluent is sold separately.

Performance characteristics

1. Sensitivity
- (1) When HISCL HBsAb C0 is analyzed, the light intensity is ≤ 7,000 counts.
- (2) When HISCL HBsAb C1 and HISCL HBsAb C0 are analyzed, the difference of light intensity is ≥30,000 counts.
2. Accuracy
- (1) When anti-HBs negative control serum is analyzed, the result is negative.
- (2) When all anti-HBs positive control sera (L, M, and H) are analyzed, the result is within the labeled concentration ±20 %.
3. Reproducibility
- When all anti-HBs positive control sera (L, M, and H) are analyzed simultaneously 10 times, the CV of each result is 15 % or less.
4. Measurement range
- 5.0 –1,000 mIU/mL

[Note 5] Counts:
Unit of light intensity on Sysmex automated immunoassay system.

[Note 6] IU:
International unit of anti-HBs concentration based on the WHO standard.

[Note 7] Anti-HBs positive control sera:
L: 10-20 mIU/mL
M: 70-150 mIU/mL
H: 600-800 mIU/mL

Limitations of the examination procedure

1. Interference
- Hemoglobin (519 mg/dL or lower), bilirubin (bilirubin F: 20.6 mg/dL or lower, bilirubin C: 21.1 mg/dL or lower), and chylomicrons (1,570 formazine turbidity units or lower) each have almost no effect on measurements.
2. Sample with turbidity or hemolysis may not be measured correctly.

Reagent preparation

All reagents are ready-to-use.

Primary sample collection, handling and storage

Human serum or plasma.

1. Plasma should be collected using EDTA or heparin as an anticoagulant. Do not use liquid anticoagulant, since it dilutes samples and causes incorrect results.
2. If samples must be stored, freeze at -20 °C or lower. Do not repeat freezing and thawing of samples, which may induce formation of particulates and cause incorrect results.
3. Fibrin-clotted samples should be centrifuged at 2,000 xg for 10 min to remove insoluble matter.


Disposal procedures

1. Incinerate used sample tubes or reagent bottles, or dispose of them as medical waste or industrial waste according to the rules stipulated for waste materials.
2. Sterilize any instruments or equipment that have come in contact with specimens using one of the following methods:
- Immerse in 0.05 % formalin solution at 37 °C for 72 hours or longer.
 - Immerse in 2 % glutaraldehyde solution for 1 hour or longer.
 - Immerse in a solution containing 0.1 % or more sodium hypochlorite for 1 hour or longer.
 - Autoclave at 121 °C for at least 1 hour.

Literature references

- (1) In-house data
- (2) Selection criterion of hepatitis virus markers in liver diseases (4th edition) Nippon Shokakibyo Gakkai Zasshi **103(12)**:1403 (2006)

Manufacturer

 **Japan Lyophilization Laboratory**
3-1-5 Matsuyama, Kiyose-shi, Tokyo 204-0022, Japan

Authorized representatives

Asia-Pacific: Sysmex Asia Pacific Pte Ltd.
9 Tampines Grande #06-18, Singapore 528735

Product information

HISCL Anti-HBs Assay Kit For 100 tests

Traceability of values assigned to calibrators

HISCL HBsAb C1-C3 have been adjusted based on WHO International Reference W1042.

Date of issue or revision

9/2018