Instruction for Use

【Product Name】

Carbonhydrate Antigen 19-9 (eCLIA)

【Order Information】

REF NO.	Package Size
0320500801	50 T
0320500802	2×50 T
0320500803	100 T
0320500804	2×100 T

【Intended Use】

It is used to detect carbohydrate antigen 19-9 (CA19-9) and to monitor the therapeutic effect of pancreatic and other digestive tract malignant tumors.

Clinical application:

Screening: Serum CA19-9 is generally not used for screening pancreatic cancer;

Auxiliary diagnosis: it is used for the auxiliary diagnosis of malignant tumors such as pancreas and bile duct, but the specificity is not strong enough. CA19-9 has no relationship with the size of pancreatic cancer, but when it is up to 10000 U/mL, there is almost peripheral metastasis[1]. Serum CA19-9 was also positive in gastric cancer, colon cancer and liver cancer. The serum levels of CA19-9 and cholangitis are different from those of benign liver cirrhosis and cholangitis[2-4]. 3% to 7% of the patients had Lewis antigen negative blood group structure and did not express CA19-9. Therefore, CA19-9 test results of these patients were often negative[5].

Prognosis evaluation: Serum CA19-9 combined with clinical data can be used as a comprehensive prognostic indicator for pancreatic cancer.

Efficacy and recurrence monitoring: Serum CA19-9, together with imaging examination, can be used to monitor the therapeutic effect of radiotherapy and chemotherapy for pancreatic cancer. The continuous increase of CA19-9 concentration indicates disease progression[6-7]. Serum CA19-9, together with imaging examination, can be used for follow-up and recurrence monitoring of pancreatic cancer after surgical resection. The patients were followed up every 3 months in the first year, every 6 months in the second to third years, and once a year after the operation.

【Principles of the testing method】

Sandwich principle. Total duration of assay: 9 minutes.

The Carbonhydrate Antigen 19-9 (CA19-9) Assay Kit adopts the double-site sandwich electrochemiluminescence method. CA19-9 in the sample, biotinylated CA19-9 antibody and the ruthenium (Ru) complex-labeled CA19-9 antibody form an antigen-antibody sandwich complex, which is bound to streptavidin coated magnetic particles, and then transferred to the measuring cell where the magnetic particles are captured onto the surface of the electrode, while the unbound substances are washed away. Chemiluminescence is generated after the electrode is electrified, and the generated optical signal is measured by a photomultiplier, processed by an instrument, and the concentration of CA19-9 in the sample is calculated based on the calibration curve.

【Main Components】

The reagent pack consists of MB, RA, RB, calibrators and control materials (optional), and different components and reagents of lot numbers should not be used interchangeably.

Components	Ingredients	Volume (2×50 T)	Volume (50 T)	Volume (100 T)	Volume (2×100 T)
(MB)	Streptavidin coated magnetic particles, 0.75 mg/mL; 0.1 M phosphate-buffered saline (PBS); ProClin300	2×1.8 mL	1×1.8 mL	1×3.5 mL	2×3.5 mL
(RB)	Biotinylated CA19-9 antibody, 3.0 mg/L; 50 mM HEPES; ProClin300	2×3.8 mL	1×3.8 mL	1×7.5 mL	2×7.5 mL
(RA)	Ru complex-labeled CA19-9 antibody, 1.5 mg/L; 50 mM HEPES; ProClin300	2×3.8 mL	1×3.8 mL	1×7.5 mL	2×7.5 mL
Calibrator	CA19-9 antigen, 100 mM PBS pH7.4, ProClin 300	High: 2×1.0 mL Low: 2×1.0 mL	High: 1×1.0 mL Low: 1×1.0 mL		
Control Material (Optional)	CA19-9 antigen, 100 mM PBS pH7.4, ProClin 300	High: 2×1.0 mL Low: 2×1.0 mL	High: 1×1.0 mL Low: 1×1.0 mL		

The assignment process of supporting calibrators in this pack is strictly implemented with reference to ISO 17511:2020, which can be traced to the Roche ELecsys CA19-9.

Refer to the quality control card for the target value and range of the quality controls.

Supporting instruments and materials required but not provided in this pack (provided by Lifotronic).

eCL8000, eCL8000i, eCL8000p, eCL8000x:

- Auffer 480 mL
- Buffer 480 mL
- Concentrated washing buffer
- Assay cup
- Sample diluent

eCL9000, eCL9000i, eCL9600, eCL9900, eCL9900i:

- Auffer 2 L
- Buffer 2 L
- Concentrated washing buffer
- Assay cup
- Disposable sampling head
- Preconditioning bath
- Waste bags
- Sample diluent

【Storage and Shelf Life】

Unopened reagent kit, calibrators and control materials should be placed at 2~8ºC and will be valid for 18 months.

Once opened, they can be stored at 2~8ºC for 56 days, and reagent kit can also be stored in machine (4~15ºC).

The manufacturing date is labeled on the box, kit and bottles, and the expiration time is 18 months after production.

Damaged, expired or contaminated reagents should be discarded.

【Applicable Instrument】

Automated ECL Immunoassay Analyzer: eCL8000, eCL8000i, eCL8000p, eCL8000x, eCL9000, eCL9000i, eCL9600, eCL9900, eCL9900i.

【Specimen collection, preparation and storage】

It is recommended to use human serum samples or plasma samples collected with EDTA-2K, EDTA-3K, heparin lithium blood collection tubes.

Blood samples should be collected in accordance with the standard operation of venipuncture. After the sample is completely coagulated, centrifugation should be performed to remove residual cellular substances. The sample should be free of air bubbles during testing. Lipid layer covering the sample after centrifugation should be removed. It is not recommended to use hemolyzed samples.

Samples can be stored for 3 days at room temperature (18~28ºC), 8 days at 2~8ºC, and 3 months at -15ºC or below. Samples should avoid repeated freeze-thaw cycles. Do not use the samples after three freeze-thaw cycles.

【Testing Procedure】

Testing procedure and precautions

Before testing, the system operation manual of the measuring instrument should be carefully read, so as to obtain relevant information such as system operating procedures, sample management, safety precautions and maintenance, and materials required for testing should be prepared. CA19-9 testing procedures should be called and set up in accordance with the system operating procedures.

Before using the reagent, put it into the analyzer 30 minutes in advance to automatically stir the magnetic bead particles and keep them in suspension.

Recommended environment temperature for testing: 10~30ºC; relative humidity: ≤ 80%.

The CA19-9 test adopts the double-site sandwich electrochemiluminescence method, with the total test time of 9 minutes, and the test method is as follows:

Step 1: After the user applies for testing, the system automatically aspirates 75 μL of biotinylated CA19-9 antibody, 75 μL of Ru complex-labeled CA19-9 antibody, 35 μL of streptavidin coated magnetic particles and 10 μL of sample into the reaction cup. They are automatically incubated at 37ºC for 9 min to form antigen-antibody sandwich complexes, and then the whole complexes are bound to the magnetic particles under the interaction of biotin and streptavidin.

Step 2: After incubation, the system automatically aspirates the reaction mixture into the measuring cell, the magnetic particles are captured onto the surface of electrode, while the unbound substances are washed away by buffer, and the electrodes is applied with voltage to generate chemiluminescence. The generated optical signals are measured by the photomultiplier, and the measured results are automatically determined via the calibration curve specifically generated by the instrument (this curve is obtained by performing two-point calibration for the master calibration curve obtained by reading the reagent RFID).

Calibration

Calibration should be performed using the lot-matching reagent and calibrators.

The target value of the calibrator has been written into the Radio-Frequency Identification card (RFID card) of the kit.

1. Before calibration, the target value, reagent information and master calibration curve information of the calibrator in the RFID card (Radio Frequency Identification, inductive electronic chip or proximity card) of the kit should be imported into the test system by swiping the card.
2. The supporting calibrator is tested, and the analyzer adjusts the master calibration curve according to the test results of the supporting calibrator to obtain the calibration curve tested by the current system (the analyzer can automatically interpret the validity of the calibration curve according to the adjustment results).

The test system should re-perform the calibration operation under the following conditions:

(1) When different lots of reagents are used;

(2) When the same lot of reagents has been used on the analyzer for more than 28 days;

(3) According to requirements: e.g. if the quality control result exceeds the defined limit;

(4) When changing the buffers of different lot numbers.

Quality Control procedure

In order to ensure the reliability of test results, it is recommended to test the control materials every 24 hours. It is recommended to test the control materials after each calibration, reagent lot replacement, maintenance or troubleshooting. The quality control results should all fall within the defined range. If they exceed the defined range, the reasons such as instrument status, reagents and calibrators should be investigated and the corrective measures should be taken.

Calculation

The system software can automatically calculate the analyte concentration, with the result in U/mL.

Specimen dilution

CA19-9 concentration in the sample higher than the upper limit of detection can be diluted with sample diluent.

The recommended dilution ratio is 1:10 (automatic dilution by instrument or

manual dilution). If manual dilution, the result should be multiplied by the dilution ratio. If instrument automatic dilution, the instrument will automatically calculate the result.

【Biological Reference Interval】

By analyzing the serum samples of healthy people from hospitals, it was calculated that the upper limit of the reference range of 95% is 27.4 U/mL.

Due to the differences in geography, race, gender and age, it is recommended that each laboratory determine the applicability of reference range through tests, and establish the reference range of this laboratory if necessary.

【Interpretation of results】

When interpreting the test results, it is necessary to refer to the patient's overall clinical situation, including: symptoms, medical history as well as other corresponding data and information.

【Limitations】

The test results are only for clinical reference and cannot be used alone as the basis for diagnosis or exclusion of cases.

The detection range of the kit is 0.6~1000 U/mL. If the CA19-9 concentration in the sample is lower than the lower limit of detection, the result is reported as < 0.6 U/mL; if the Ca19-9 concentration in the sample is higher than the upper limit of detection, the result is reported as > 1000 U/mL (10× diluted sample is reported as > 10000 U/mL).

When jaundice (bilirubin) is ≤ 66 mg/dL, hemolysis (hemoglobin) is ≤ 2200 mg/dL, lipemia (triglyceride) is ≤ 1500 mg/dL, biotin is ≤ 20 ng/mL, and HAMA is ≤ 100 ng/mL in the sample, the interference deviation of the measured results is within ±15%. When the concentration of rheumatoid factor in the sample is ≤ 1500 IU/mL, the relative recovery of the test results is between 85% and 115%.

When the sample contains adriamycin (7 μg/mL), methotrexate (8.5 μg/mL), cyclophosphamide (210 μg/mL), 5-fluorouracil (220 μg/mL), cisplatin (57 μg/mL), gemcitabine (382 μg/mL) Leucocorin (115 μg/mL) and dexamethasone (10 μg/mL), the interference deviation of the measured results is within ±15%.

Samples of other tumor markers containing 1000 ng/mL AFP, 1000 U/mL CA125, 100 U/mL CA15-3, 1000 ng/mL CEA, 100 ng/mL PSA and 1000 ng/mL Ferr are tested, and the CA19-9 measured results are not greater than 10 U/mL.

When the CA19-9 concentration reaches 350000 U/mL, the test results are not be affected by the hook effect.

【Analytical Performance】

Lower limit of measurement

Limit of Blank (LoB) ≤ 0.6 U/mL.

Accuracy

The relative bias of measuring Accuracy control materials should fall between 85% and 115%.

Linearity

Within the range of 0.6 U/mL~1000 U/mL, the correlation coefficient (r) of the kit should not be less than 0.9900.

Within-run imprecision (repeatability)

The coefficient of variation (CV) is less than 5.0%.

Between-lot imprecision

The coefficient of variation (CV) is less than 10.0%.

Homogeneity of calibrators and control materials

In-bottle homogeneity: coefficient of variation (CV) ≤ 5%.

Accuracy of calibrators

The supporting calibrators of the kit are tested, the relative deviation of their test results is within ±15%.

Measured values of quality control

The measured results of supporting assigned quality control of the kit should be within the quality control range.

【Precaution and Warning】

This kit is only used for in vitro diagnosis;

When using this kit, the relevant operation precautions of the laboratory must be observed;

The test results of this kit can only be used as clinical reference, the patient's clinical evaluation should be based on the patient's clinical symptoms/signs, medical history, other laboratory test results and treatment response, etc.;

Due to methodological reasons or antibody specificity, the results of the same sample tested with reagents of different manufacturers may be different. Therefore, results obtained with different kits should not be directly compared, so as to prevent wrong medical interpretation; it is recommended that the Laboratory Department indicate the characteristics of the reagents in the test report issued to the clinician. If the reagent type is changed during series monitoring, continuous testing should be conducted, and the results should be compared with the original reagent results in parallel to re-determine the baseline value;

This product contains animal-derived substances, and may have potential biological risks. All samples and reaction wastes should be treated as the source of infection, and all wastes must be disposed of in accordance with local regulations.

【Symbol】

Symbol	Title of Symbol	Symbol	Title of Symbol
	Manufacturer		Consult instructions for use
EC REP	Authorized representative in the European Community	IVD	In vitro diagnostic medical device
	Use-by date	CE	Indicates this device is in compliance with Europe Directive
LOT	Batch code	Σ	Contains sufficient for<*n*>tests
SN	Serial number		Biological risks
	Temperature limitation		This way up
REF	Catalogue number		

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【Version and Revision】

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