This package insert follows the Pharmaceuticals, Medical devices and Other Therapeutic Products Act of Japan.

In Vitro Diagnostics
Marketing Notification No. 13A2X00197218045

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This package insert must be read carefully prior to use.

Cholesterol assay kit
(Classification No.: 30159000)

QUALIGENT CHO

General Precautions

1. This product is for in vitro diagnostic use, and must not be used for any other purposes.
2. Clinicians should make a comprehensive clinical decision based on assay results in conjunction with clinical symptoms and other examination results.
3. This product should be used only as directed in this package insert. Reliability of results cannot be guaranteed if there are any deviations from the instructions in this package insert.
4. If the reagent accidentally comes in contact with eyes and/or mouth, rinse immediately with ample water as first aid, and consult the doctor if required.
5. Carefully read the operating instructions for each type of automated analyzers prior to using this product. Parameters for each type of analyzers are available, and can be requested from SEKISUI MEDICAL CO., LTD. if required.
6. Perform a quality control test prior to assay to ensure accuracy.

Description (Kit Components) **

Component: Ingredients
CHO Enzyme Solution 1:
4-Aminoantipyrine
Cholesterol esterase
Peroxidase
CHO Enzyme Solution 2:
Cholesterol oxidase (microbial origin)
N-ethyl-N-sulfobutyl-m-toluidine sodium

Intended Use

Measurement of total cholesterol in serum or plasma

Cholesterol in serum or plasma is derived from dietary intake and is also synthesized in the liver. Measurement of cholesterol is important as an index of abnormal lipid metabolism and is also considered to be useful for the diagnosis of hepatic dysfunction and metabolic disease.

Assay Principle

1. Assay Principle

Cholesterol exists in samples as both the esterified and free types. Esterified cholesterol is converted to free cholesterol by the action of cholesterol esterase (CE). Then free cholesterol is oxidized by cholesterol oxidase (COD) to form hydrogen peroxide. Hydrogen peroxide causes oxidative condensation of 4-aminoantipyrine and N-ethyl-N-sulfobutyl-m-toluidine (ESBmT) in the presence of peroxidase (POD) to form a red-purple color. Total cholesterol content is determined by measuring the absorbance of the sample. The influence of ascorbic acid is blocked by ascorbate oxidase.

$$\text{Esterified cholesterol} \xrightarrow{\text{CE}} \text{Free cholesterol} + \text{Fatty acid}$$

$$\text{Free cholesterol} \xrightarrow{\text{COD}} \Delta^4\text{-cholestenone} + H_2O_2$$

$$H_2O_2 + \text{ESBmT} + \text{4-Aminoantipyrine} \xrightarrow{\text{POD}} \text{Red-purple color}$$

2. Features

1) Assay results are hardly affected by cross contamination.
2) Bilirubin, hemolysis, ascorbic acid and chyle have minimal effects on results.

Procedural Precautions *

1. Properties of Samples and Sampling Methods

1) Samples
Serum and plasma (heparin plasma, EDTA plasma and citrated plasma) may be used.
2) Storage of samples[1)]
If the isolated serum or plasma sample cannot be tested on the same day, specimens should be stored as follows:
2–10°C: for tests within 1 week
≤ -20°C: for tests after more than 1 week
Bring samples to room temperature (15–30°C) before use.

2. Interfering substances

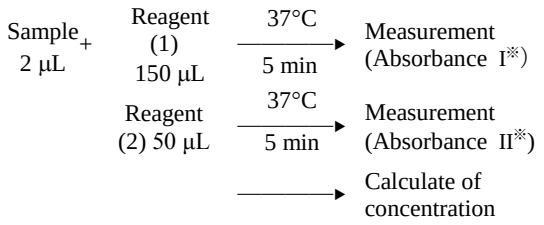
Assay results are not affected by free bilirubin (up to 20 mg/dL), conjugated bilirubin (up to 20 mg/dL), hemoglobin (up to 500 mg/dL), ascorbic acid (up to 50 mg/dL), formazin turbidity (up to 3000 FTU), or Intralipos (up to 5%).

3. Others

1) Always use Cholestest N Calibrator for calibration.
However, for Hitachi LABOSPECT 008 Automated Analyzer, use QUALIGENT N Calibrator (manufactured by SEKISUI MEDICAL) for calibration.
2) Precautions for assay range
If the concentration of sample exceeds assay range, dilute the sample with saline and repeat the measurement.

Dosage/Administration (Assay Procedure) * *

1. Preparation of reagents
Reagent (1): CHO Enzyme Solution 1 is ready to use.
Reagent (2): CHO Enzyme Solution 2 is ready to use.
2. Assay Procedure
This product is compatible with Hitachi 9000 series and LABOSPECT series automated analyzers. Assay procedure is indicated below.



※ Absorbance I and II: The absorbance difference between 800 nm and 600 nm.

Calibration material: Cholestest N Calibrator

Use QUALIGENT N Calibrator (manufactured by SEKISUI MEDICAL) for Hitachi LABOSPECT 008 Automated Analyzer for calibration. (Manufacture's assigned values)

Reagent blank: Purified water or saline

Assessment of Assay Results **

1. Reference standard range[2)]
142–248 mg/dL

2. Diagnostic criterion[3)]
Hypercholesterolemia: ≥ 220 mg/dL

3. There may be reactions or interfering reactions with non-target substances. If assay results appear to be unreliable, repeat the measurement (if necessary, after dilution) or try another analytical methods.

Performance **

1. Sensitivity
1) Reagent blank: absorbance being equal to or lower than 0.05
2) Sensitivity: The absorbance is 0.40–0.72 per 300 mg/dL of cholesterol.

2. Accuracy: 95–105% of the expected assay value

3. Within-run Reproducibility:
Coefficient of variation ≤ 3%
(Test methods used for 1.-3. are in-house methods.)

4. Measurement Range[4)]: (On a Hitachi 9000 series automated analyzer)
5–1000 mg/dL

5. Correlation[4)]
1) Serum N=70 r=0.997 y=1.01x–2.06
Control method: Approved in vitro diagnostic (enzymatic method)
2) Plasma N=50 r=0.999 y=1.00x –1.26
Control method: Approved in vitro diagnostic (enzymatic method)

6. Standard Material
SRM911（NIST）

Precautions for Use or Handling * *

1. Precautions for Handling (to Ensure Safety)
1) All samples used in the test should be handled as a material possibly infected with HIV, HBV, HCV, or other viruses. To prevent infection, use disposable gloves and avoid mouth pipetting during the test.
2) Cholestest N Calibrator and QUALIGENT N Calibrator for LABOSPECT 008 contain human-derived components that have been shown to be negative for HBs antigens, HIV antibodies (AIDS virus antibodies), and HCV antibodies. However, these reagents (as well as the samples) should be considered potentially infectious and handled with great care.
3) Proclin 300, which possesses skin-irritative potential, is added as an antiseptic agent in the CHO Enzyme Solution 1. Therefore, if the reagent comes in contact with skin or clothes, rinse immediately with ample water, and consult the doctor if skin irritation develops.
4) Sodium azide is added as an antiseptic agent in the CHO Enzyme Solution 2. Therefore, if the reagent comes in accidentally contact with eyes, mouth or skin, rinse immediately with ample water as first aid, and consult the doctor if required.

2. Precautions for use
1) This product should be stored as directed, without freezing. Freezing can deteriorate the reagents, which can produce inaccurate results. Therefore, avoid using the reagents which have been previously frozen.
2) Do not use expired reagents. Use of such reagents cannot guarantee the reliability of measurement values.
3) Do not perform the assay under direct sunlight

3. Precautions for Disposal
1) Before disposal, used samples and their containers must be immersed in sodium hypochlorite solution at a concentration of greater than 0.1% for longer than 1 hour or autoclaved at 121ºC for 20 minutes.
2) To prevent infections from spilled samples or solutions containing samples, wipe the spilled area thoroughly with disinfectants such as sodium hypochlorite solution at a concentration of greater than 0.1%.
3) The reagents and treated samples should be discarded as medical waste or industrial waste according to the waste disposal regulations.
4) The reagents should be disposed of in accordance with the Water Pollution Control act or related regulations.
5) Sodium azide is added as an antiseptic agent in the CHO Enzyme Solution 2. It can react with lead or copper pipes to produce the highly explosive metal azide. Therefore, the reagent should be flushed with large amounts of water when disposing.

4. Other precautions
1) Do not use the containers for other purposes.
2) Do not take apart the reagent cartridge before.

Storage and Shelf Life **

1. Storage temperature: 2–10°C

2. Shelf life: 18 months from the date of manufacture
(The expiration date is printed on the outer package.)

Packaging * *

Name	Package contents		
QUALIGENT CHO	Set (Cassette for Hitachi 9000 series)	CHO Enzyme Solution 1 1 × 22.5 mL	× 2
		CHO Enzyme Solution 2 1 × 7.5 mL	
	L set (Set for Hitachi LABOSPECT series)	CHO Enzyme Solution 1 1 × 46 mL	× 2
		CHO Enzyme Solution 2 1 × 16 mL	

References * *

1) Sasaki M. et al.: Sampling of constituents of the human body, 246, Kodansha, 1972.
2) Kanai M. (supervising editor): Kanai's manual of clinical laboratory medicine. 34th ed. 512, Kanehara Shuppan, 2015.
3) Japan Atherosclerosis Society, ed. Japan Atherosclerosis Society (JAS) Guidelines for Diagnosis and Treatment of Atherosclerotic Cardiovascular Diseases, 2002, 5.
4) In house data, SEKISUI MEDICAL CO., LTD.

Contact

SEKISUI MEDICAL CO., LTD.
international@sekisui.com

Manufacturer * *

SEKISUI MEDICAL CO., LTD.
1-3, Nihonbashi 2-chome, Chuo-ku, Tokyo, Japan