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

In Vitro Diagnostics
Marketing Notification No. 13A2X00197219005

** Revised: January 2017 (4th edition)
* Revised: August 2011 (3rd edition)

This package insert must be read carefully prior to use.

Alanine aminotransferase assay kit
(Classification No.: 38556000)

QUALIGENT ALT-L

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

ALT-L Enzyme Solution 1:
Lactate dehydrogenase
Nicotinamide adenine dinucleotide (reduced form)
L-alanine

ALT-L Substrate Solution 2:
L-alanine
α-Ketoglutaric acid
2-Amino-2-hydroxymethyl-1, 3-propanediol buffer

Intended Use

Measurement of alanine aminotransferase (ALT) in serum or plasma

ALT is an enzyme (aminotransferase) that catalyzes the transfer of an amino group between amino acids and α-keto acids. It is widely distributed in the myocardium, liver, and brain.
Blood ALT activity is markedly elevated in hepatic and biliary tract diseases, particularly acute hepatitis. It is also elevated in myocardial infarction.

Assay Principle

1. Assay Principle

ALT catalyzes a reaction producing pyruvic acid and glutamic acid from L-alanine and α-ketoglutaric acid. The resulting pyruvic acid is converted to lactic acid and NAD by lactate dehydrogenase (LD) in the presence of NADH. The reaction also converts NADH to NAD, and its absorbance at 340 nm decreases.

ALT activity is determined by measuring the velocity of the decrease in NADH.

$$\text{L-alanine} + \alpha\text{-Ketoglutaric acid} \xrightarrow{\text{ALT}} \text{Pyruvic acid} + \text{Glutamic acid}$$

$$\text{Pyruvic acid} + \text{NADH} \xrightarrow{\text{LD}} \text{Lactic acid} + \text{NAD}$$

2. Features

Final concentration of the principal ingredient matches with the final concentration stipulated in the JSCC Recommendation for Enzyme Activities.

Procedural Precautions

1. Properties of Samples and Sampling Methods

1) Samples

Serum and plasma (heparin plasma, EDTA plasma, citrated plasma and NaF-EDTA plasma) may be used.

2) Storage of samples

If the isolated serum or plasma sample cannot be tested on the same day, specimens should be stored as follows[7]:

2–10°C: for tests within 2 weeks
≤ -20°C: for tests after more than 2 weeks

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), formazin turbidity (up to 2500 FTU), or ascorbic acid (up to 50 mg/dL).

3. Others

1) Always use Enzyme Calibrator Plus "Daiichi" for calibration.

2) Precautions for assay range

If the activity of sample exceeds assay range, dilute the sample with saline and repeat the measurement.

Dosage/Administration (Assay Procedure) **

1. Preparation of reagents

Reagent (1): ALT-L Enzyme Solution 1 is ready to use.
Reagent (2): ALT-L Substrate 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.

Sample 5.5 μL + Reagent (1) 100 μL → (37°C, 5 min) → Reagent (2) 50 μL → (37°C, 74–299 sec) → Measurement (Absorbance※) → Calculation of activity

※ Absorbance: The difference in absorbance between 546 nm and 340 nm
Calibration material: Enzyme Calibrator Plus "Daiichi" (Manufacture's assigned value)
Reagent blank: Purified water or saline

Assessment of Assay Results **

1. Reference standard range[8)]
Male: 10–42 U/L
Female: 7–24 U/L (≤ 45 years)
9–32 U/L (> 45 years)

2. 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: change in absorbance being equal to or lower than 0.012/min
2) Sensitivity: Absorbance variation is 0.017–0.024/min per 100 U/L of ALT activity.

2. Accuracy: 90–110% of the expected assay value

3. Within-run Reproducibility:
Coefficient of variation ≤5%
(Test methods used for 1. –3. are in-house methods.)

4. Analysis range[9)]: (On Hitachi 9000 series automated analyzer)
3–1500 U/L

5. Correlation [9)]
1) Serum N=50 r=0.999 y=0.97x –0.04
Control method: using an already approved in vitro diagnostic product with method in line with the JSCC Recommendation for Enzyme Activities.
2) Plasma N=60 r=0.999 y=0.98x +0.54
Control method: using an already approved in vitro diagnostic product with method in line with the JSCC Recommendation for Enzyme Activities.

6. Standard Material
Enzyme Calibrator Plus, the calibration material used for this product, is in line with the Japanese Standard for Certified Enzyme Reference Materials

Precautions for Use and 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) Sodium azide is added as an antiseptic agent in the ALT-L Substrate 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 ALT-L Substrate 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 use.

Storage and Shelf Life **

1. Storage temperature: 2–10°C
2. Shelf life: 9 months from the date of manufacture
(The expiration date is printed on the outer package.)

Packaging **

Name	Package		
QUALIGENT ALT-L	Set (Cassette for Hitachi 9000 series)	ALT-L Enzyme Solution 1 1×15.0 mL	×2
		ALT-L Substrate Solution 2 1 × 7.5 mL	
	L set (Set for Hitachi LABOSPECT series)	ALT-L Enzyme Solution 1 1 × 40 mL	×2
		ALT-L Substrate Solution 2 1 × 20 mL	

Constituent reagents are available in other configurations. For further details please contact SEKISUI MEDICAL CO., LTD.

References **

1) Japan Society of Clinical Chemistry: Jpn J Clin Chem, 18, 250, 1989.
2) Japan Society of Clinical Chemistry: Jpn J Clin

Chem, 18, 255, 1989.
3) Kotani K. et al.: J Clin Lab Inst Reag, 16, 153, 1993.
4) Kotani K. et al.: J Jpn Soc Gastroenterol, 91, 154, 1994.
5) Osawa S.: Med Tech, 20, 1022, 1992.
6) Japan Society of Clinical Chemistry: Jpn J Clin Chem, 25, 135, 1996.
7) Sasaki M. et al.: Sampling of constituents of the human body, 211, Kodansha, 1972.
8) Kanai M. (supervising editor): Kanai's manual of clinical laboratory medicine. 34th ed. 579, Kanehara Shuppan, 2015.
9) 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