DXC (ALT) ALANINE AMINOTRANSFERASE

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PURPOSE

To provide instructions for the quantitative determination of alanine aminotransferase on the DXC 600/800.

PRINCIPLE

ALT reagent, when used in conjunction with UniCel® DxC 600/800 System(s), is intended for the quantitative determination of Alanine Aminotransferase activity in human serum or plasma.

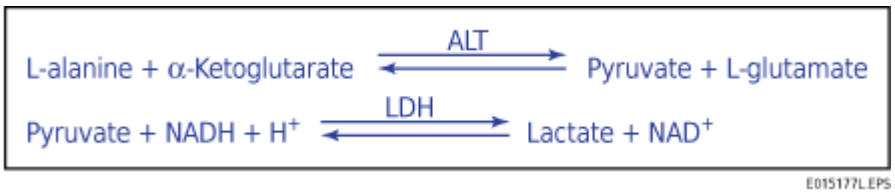
BACKGROUND

Clinical Significance

Alanine aminotransferase measurements are used in the diagnosis and treatment of certain liver diseases (e.g., viral hepatitis and cirrhosis) and heart diseases.

Methodology

ALT reagent is used to measure analyte activity by a kinetic rate method. In the reaction, alanine aminotransferase catalyzes the reversible transamination of L-alanine and alpha-ketoglutarate to pyruvate and L-glutamate. The pyruvate is then reduced to lactate in the presence of lactate dehydrogenase (LDH) with the concurrent oxidation of reduced β-nicotinamide adenine dinucleotide (NADH) to β-nicotinamide adenine dinucleotide (NAD).
The SYNCHRON® System(s) automatically proportions the appropriate sample and reagent volumes into the cuvette. The ratio used is one part sample to 11 parts reagent. The system monitors the change in absorbance at 340 nanometers. This change in absorbance is directly proportional to the activity of ALT in the sample and is used by the System to calculate and express the ALT activity.



RELATED DOCUMENTS

R-PO-CH0810	Quality Control Program General Laboratory
R-PO-CH0809	Quality Control Westgard Rules Statistics
R-PR-AD0540	Specimen Rejection/Cancellation Protocol
J-F-CH0820	DXC 800 Controls
M-F-CH0820	Chemistry Controls
J-F-CH0826	DXC 800 Calibrators
M-F-CH0826	Chemistry Calibrators

M-F-CH1940	DXC 600 (AMR) Analytical Measurement Range
J-F-CH1940	DXC 800 (AMR) Analytical Measurement Range
R-W-CH0815	DXC Reagent Lot to Lot Correlations
R-F-CH0814	Lot-to-Lot Correlation

SPECIMEN

Type of Specimen

Biological fluid samples should be collected in the same manner routinely used for any laboratory test. Freshly drawn serum or plasma is the specimen of choice. Acceptable anticoagulants are listed in PROCEDURAL NOTES section of this chemistry information sheet. Whole blood is not recommended for use as a sample.

Specimen Storage and Stability

1. Tubes of blood are to be kept closed at all times and in a vertical position. It is recommended that the serum or plasma be physically separated from contact with cells within two hours from the time of collection.

2. Separated serum or plasma should not remain at room temperature longer than 8 hours. If assays are not completed within 8 hours, serum or plasma should be stored at +2°C to +8°C. If assays are not completed within 48 hours, or the separated sample is to be stored beyond 48 hours, samples should be frozen at -15°C to -20°C. Frozen samples should be thawed only once. Analyte deterioration may occur in samples that are repeatedly frozen and thawed.

3. Refer to references (4,5) for additional information on the effects of preanalytical variables on sample storage and stability. Each laboratory should determine if the recommended requirements are appropriate.

Sample Type	Volume	Sample Stability
Plasma/Serum	0.5mL	• Separate serum from cells within 2 hours • Room Temp 8 hours • Refrigerated 48 hours • Frozen 3 months

Criteria for Unacceptable Specimens

See Specimen Rejection/Cancellation Protocol

Sample Volume

A filled 0.5 mL sample cup is the optimum volume. For optimum primary sample tube volumes in primary tube samples and minimum volumes, refer to the Primary Tube Sample Template for your system.

REAGENTS

Contents

Each kit contains the following items:
Two Alanine Aminotransferase Reagent Cartridges (2 x 200 tests) or (2 x 400 tests and 2 bottles of ALT [A-reagent])

Volume per Test	
Sample Volume	12 µL
Ordac Sample Volume	3 µL
Total Reagent Volume	250 µL
Cartridge Volumes	A 242 µL B 8 µL C – –

Reactive Ingredients	
α-Ketoglutarate	16 mmol/L
Lactate dehydrogenase (LD)	>2300 IU/L
L-Alanine	500 mmol/L
Tris buffer	97 mmol/L
NADH	0.18 mmol/L

Also non-reactive chemicals necessary for optimal system performance.

Reagent Preparation

For P/N 442620 (200 tests): Transfer all the contents of the smallest reagent compartment (C) into the largest reagent compartment (A).

For P/N 476826 (400 tests): Transfer all the contents of one ALT (A-reagent) bottle into the largest reagent compartment (A).

Replace cartridge caps and gently invert cartridge several times to ensure adequate mixing.

Acceptable Reagent Performance

The acceptability of a reagent is determined by ensuring that quality control results are within your facility`s acceptance criteria.

NOTE: New lots of reagent require lot to lot correlation studies. Refer to Related Documents section for related work instructions/forms.

Reagent Storage and Stability

ALT reagent when stored unopened at +2°C to +8°C will obtain the shelf-life indicated on the cartridge label. Once prepared, the reagent is stable for 30 days at +2°C to +8°C. Do not use beyond the manufacturer's expiration date. DO NOT FREEZE.

CALIBRATION

Calibrator Required

Calibration is not required.

Traceability

This measurand (analyte) is traceable to the manufacturer's selected Measurement Procedure as described in the Methodology section.

QUALITY CONTROL

See Related Documents J-F-CH0820 DXC 800 Controls & M-F-CH0820 Chemistry Controls

STEPS

1. If necessary prepare reagent as defined in the Reagent Preparation section of this chemistry information sheet and load the reagent onto the system.

2. Program controls for analysis.

3. After loading controls onto the system, follow the protocols for system operation**.** To load samples manually refer to the FHS DXC Series Manual Sample Programming procedure. For detailed testing procedures, refer to the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual.

CALCULATIONS

SYNCHRON® System(s) perform all calculations internally to produce the final reported result. The system will calculate the final result for sample dilutions made by the operator when the dilution factor is entered into the system during sample programming.

ANTICOAGULANT TEST RESULTS

1. If plasma is the sample of choice, the following anticoagulants were found to be compatible with this method based on a study of 20 healthy volunteers:

Anticoagulant	Level Tested for In Vitro Interference
Ammonium Heparin	14 Units/mL
Lithium Heparin	14 Units/mL
Sodium Heparin	14 Units/mL

2. The following anticoagulant was found to be incompatible with this method:

Anticoagulant	Level Tested for In Vitro Interference
Potassium Oxalate	2 Units/mL
Sodium Fluoride	2.5 Units/mL

PERFORMANCE CHARACTERISTICS

Reference Range

Sample Type	Conventional Units
Serum or Plasma	10 –65 U/L

Analytic Range

The SYNCHRON® System(s) method for the determination of Alanine Aminotransferase provides the following analytical range:

Sample Type	Conventional Units
Serum or Plasma	5 – 400 U/L
Serum or Plasma (Ordac)	350-2600 IU/L

Samples with activities exceeding the high end of the analytical range should be rerun with ORDAC enabled or diluted with saline and reanalyzed. The appropriate dilution factor should be applied to the reported result.

Reporting results outside of analytical range

Lower limit of detection	5 IU/L	Results below 5; Report as <5 IU/L (See Limitations below for "OIR LO" results)
Upper limit of detection	2600 IU/L	Results >2600 should be diluted with 0.9% saline, reanalyzed and dilution factor applied. The maximum allowable dilution is X5. Results >13,000 are reported as >13,000 IU/L.

Sensitivity

Sensitivity is defined as the lowest measurable concentration which can be distinguished from zero with 95% confidence. Sensitivity for ALT determination is 5 IU/L.

LIMITATIONS

A sample with a suspected "true" low result exceeding the analytical range that suppresses the result as "OIR LO" can be confirmed by adding a measured volume of the test sample to an equal volume of material with an assigned value or concentration (x2 dilution).The low result is confirmed when the diluted test sample is within the assigned concentration.

Samples with extremely high enzyme activity (>12,000 IU/L) ("worst case scenario") may consume all of the NADH substrate before the first absorbance measurement is taken after sample addition. These samples can report either very low enzyme activities or suppress the result as "OIR LO". These samples should be diluted 1:20 with saline and rerun.

Interferences

1. The following substances were tested for interference with this methodology:

Substance	Source	Level Tested	Observed Effect
Hemolysis	RBC Hemolysate	INDEX of 1	AVOID HEMOLYSIS
Bilirubin	Bovine	30 mg/dL INDEX of 20	No significant interference (within ± 6 IU/L or 7%)
Lipemia	Intralipid	320 mg/dL INDEX of 8 Airfuge recommended	No significant interference (within ± 6 IU/L or 7%)

2. Samples showing evidence of hemolysis should not be used. Hemolysis may cause falsely elevated results.

3. Refer to References (10,11,5) for other interferences caused by drugs, disease and preanalytical variables.

ADDITIONAL INFORMATION

For more detailed information on UniCel DxC Systems, refer to the appropriate system manual.

REFERENCES

1. Kamen, A., Wroblewski, F., LaDue, J. E., *J. Clin. Inv.*, 34 :126 133 (1955).
2. Henry, R. J., et al., *Amer. J. Clin. Path.*, 34:381 (1960).
3. Tietz, N. W., "Specimen Collection and Processing; Sources of Biological Variation", *Textbook of Clinical Chemistry*, 2nd Edition, W. B. Saunders, Philadelphia, PA (1994).
4. National Committee for Clinical Laboratory Standards, *Procedures for the Handling and Processing of Blood Specimens*, Approved Guideline, NCCLS publication H18-A, Villanova, PA (1995).
5. Young, D. S., *Effects of Preanalytical Variables on Clinical Laboratory Tests*, 2nd Edition, AACC Press, Washington, D. C. (1997).
6. Tietz, N. W., *Clinical Guide to Laboratory Tests*, 3rd Edition, W. B. Saunders, Philadelphia, PA (1995).
7. National Committee for Clinical Laboratory Standards, *How to Define, Determine, and Utilize Reference Intervals in the Clinical Laboratory*, Approved Guideline, NCCLS publication C28-A, Villanova, PA (1995).
8. Tietz, N. W., ed., *Fundamentals of Clinical Chemistry*, 3rd Edition, W. B. Saunders, Philadelphia, PA (1987).
9. Henry, J. B., *Clinical Diagnosis and Management by Laboratory Methods*, 18th Edition, W. B. Saunders Company, Philadelphia, PA (1991).
10. Young, D. S., *Effects of Drugs on Clinical Laboratory Tests*, 4th Edition, AACC Press, Washington, D. C. (1995).
11. Friedman, R. B., Young, D. S., *Effects of Disease on Clinical Laboratory Tests*, 3rd Edition, AACC Press, Washington, D.C. (1997).
12. National Committee for Clinical Laboratory Standards, *Method Comparison and Bias Estimation Using Patient Samples*, Approved Guideline, NCCLS publication EP9-A, Villanova, PA (1995).
13. National Committee for Clinical Laboratory Standards, *Precision Performance of Clinical Chemistry Devices*, Tentative Guideline, 2nd Edition, NCCLS publication EP5-T2, Villanova, PA (1992).