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CE CORMAY

Liquick Cor - ASAT

Kit name	(EN) Cat. No
Liquick Cor-ASAT mini	1-290
Liquick Cor-ASAT 30	1-222
Liquick Cor-ASAT 60	1-214
Liquick Cor-ASAT 120	1-215
Liquick Cor-ASAT 500	1-313

INTENDED USE
Diagnostic kit for determination of aspartate aminotransferase activity intended to use for manual assay (Sample Start and Reagent Start method) and in several automatic analysers.
The reagents must be used only for *in vitro* diagnostic, by suitably qualified laboratory personnel, only for the intended purpose, under appropriate laboratory conditions.

INTRODUCTION
Aspartate aminotransferase (ASAT, AST, GOT) is an enzyme participated in amino acids metabolism. ASAT is found in all tissues but particularly high level of ASAT is observed in heart muscle, skeletal muscle, liver and kidney. This is why elevated ASAT serum level is marker of myocardial infarction and kidney, liver or skeletal muscle injury.

METHOD PRINCIPLE
Optimized, modified method according to International Federation of Clinical Chemistry (IFCC), without pyridoxal phosphate.

L-aspartate + 2-oxoglutarate $\xleftrightarrow{ASAT}$ oxalacetate + L-glutamate

oxalacetate + NADH + H^+ $\xleftrightarrow{MDH}$ malate + NAD^+

The rate of absorbance changing at λ=340 nm is directly proportional to aspartate aminotransferase activity.

REAGENTS
Package

	Liquick Cor -ASAT mini	Liquick Cor -ASAT 30	Liquick Cor - ASAT 60
1-ASAT	2 x 24 ml	5 x 24 ml	5 x 48 ml
2-ASAT	1 x 12 ml	1 x 30 ml	1 x 60 ml

	Liquick Cor - ASAT 120	Liquick Cor - ASAT 500
1-ASAT	5 x 96 ml	3 x 400 ml
2-ASAT	1 x 120 ml	1 x 300 ml

The reagents when stored at 2-8°C are stable up to expiry date printed on the package. The reagents stored on board of the analyzer at 2-10°C are stable for 12 weeks (Biolis 24i Premium).

Working reagent preparation and stability
Assay can be performed with use of separate 1-ASAT and 2-ASAT reagents or with use of working reagent. For working reagent preparation mix gently 4 parts of 1-ASAT with 1 part of 2-ASAT. Avoid foaming!

Stability of working reagent: 4 weeks at 2-8°C
5 days at 15-25°C

Concentrations in the test
1-Reagent

Tris buffer, pH 7,7	≤ 120 mmol/l
L-aspartate	≤ 360 mmol/l
MDH	≤ 1.4 U/ml
LDH	≤ 2.3 U/ml
stabilzer, preservative	

2-Reagent

2-oxoglutarate	≤ 74 mmol/l
NADH	≤ 1.7 mmol/l
buffer	
preservatives	

Warnings and notes
- Protect from direct sunlight and avoid contamination!
- The reagents are usable when the absorbance of the working reagent is higher than 1.400 (read against distilled water, wavelength λ=340 nm, cuvette l = 1 cm, at temp. 25°C).
- Please refer to the MSDS for detailed information concerning safe storage and use of the product.
- 1-Reagent meeting the criteria for classification in accordance with Regulation (EC) No 1272/2008

Warning

H315 Causes skin irritation.
H319 Causes serious eye irritation.
P280 Wear protective gloves/protective clothing/eye protection/face protection.
P302+P352 IF ON SKIN: Wash with plenty of soap and water.
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

ADDITIONAL EQUIPMENT
- automatic analyzer or photometer able to read at 340 nm (Hg 334nm, 365nm);
- thermostat at 37ºC;
- general laboratory equipment;

SPECIMEN
Serum, heparinized or EDTA plasma free from hemolysis. Do not use heparine ammonium salt.
Hemolysis should be avoided, since ASAT activity in erythrocytes is 10 times higher than in normal serum. Do not freeze the samples. ASAT activity remains stable in specimen up to 1 day at 15-25ºC or up to 4 days at 2-8ºC. Nevertheless it is recommended to perform the assay with freshly collected samples!

PROCEDURE
Applications for analyzers are available on request.

Manual procedure

wavelength	340 nm (Hg 334 nm, 365 nm)
temperature	37°C
cuvette	1 cm

Sample Start method
Pipette into the cuvette:

working reagent	1000 µl

Bring up to the temperature of determination 37°C for 10 minutes. Then add:

sample	100 µl

Mix and incubate at adequate temperature. After about 1 min. read the absorbance against air or water. Repeat the reading after exactly 1, 2 and 3 minutes. Calculate the mean absorbance change per minute (ΔA/min.).

Calculation
ASAT activity [U/l] = ΔA/min. x F

F value depends on the used wavelength:

λ	334 nm	340 nm	365 nm
F	1979	1939	4800

Reagent Start method
The determination can be also performed with use of separate 1-ASAT and 2-ASAT reagents.

Pipette into the cuvette:

1-ASAT	1000 µl

Bring up to the temperature of determination. Then add:

sample	100 µl

Mix well, incubate for 5 min. Then add:

2-ASAT	250 µl

Mix well; perform measurement as described for Sample Start method.

Calculation
ASAT activity [U/l] = ΔA/min. x F

F value depends on the used wavelength:

λ	334 nm	340 nm	365 nm
F	2670	2441	5922

REFERENCE VALUES [6]

serum / plasma	37ºC	
female	up to 31 U/l	up to 0.518 µkat/l
male	up to 37 U/l	up to 0.618 µkat/l

It is recommended for each laboratory to establish its own reference ranges for local population.

QUALITY CONTROL
For internal quality control it is recommended to use with each batch of samples, the CORMAY SERUM HN (Cat. No 5-172) and CORMAY SERUM HP (Cat. No 5-173).
For the calibration of manual assay the CORMAY MULTICALIBRATOR LEVEL 2 (Cat. No 5-175; 5-177) is recommended.
For the calibration of automatic analysers systems the CORMAY MULTICALIBRATOR LEVEL 1 (Cat. No 5-174; 5-176) and LEVEL 2 (Cat. No 5-175; 5-177) is recommended. Deionised water should be used as a calibrator 0.
The calibration curve should be prepared every 12 weeks (Biolis 24i Premium), with change of reagent lot number or as required e.g. quality control findings outside the specified range.

PERFORMANCE CHARACTERISTICS
The following results have been obtained using Multi+ for manual assay and automatic analyser Biolis 24i Premium. Results may vary if a different instrument is used.

- **Limit of quantitation (LOQ):**
  7 U/l (0.12 µkat/l) – Multi+
  7 U/l (0.12 µkat/l) – Biolis 24i Premium.

- **Linearity:**
  up to 650 U/l (10.8 µkat/l) – Multi+
  up to 780 U/l (13 µkat/l) – Biolis 24i Premium

- **Specificity / Interferences**

Haemoglobin up to 0.63 g/dl, ascorbate up to 62 mg/l, bilirubin up to 20 mg/dl and triglycerides up to 1000 mg/dl do not interfere with the test.

- **Precision (Biolis 24i Premium)**

Repeatability (run to run) n = 20	Mean [U/l]	SD [U/l]	CV [%]
level 1	41.5	1.4	3.5
level 2	201	3.7	1.8
Reproducibility (day to day) n = 80	Mean [U/l]	SD [U/l]	CV [%]
level 1	45.2	2.2	4.8
level 2	205	2.7	1.3

- **Method comparison**

A comparison between ASAT values determined at **Multi+** (y) and at **Beckman Coulter AU680** (x) using 21 serum samples gave following results:

Sample Start method
y = 0.8974 + 2.5473 U/l;
R = 1.000 (R – correlation coefficient)

Reagent Start method
y = 0.903 + 1.3563 U/l;
R = 0.999 (R – correlation coefficient)

A comparison between ASAT values determined at **Biolis 24i Premium** (y) and at **Cobas 6000** (x) using 150 serum samples gave following results:
y = 1.0519x + 0.4975 U/l;
R = 0.999 (R – correlation coefficient)

WASTE MANAGEMENT
Please refer to local legal requirements.

LITERATURE
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