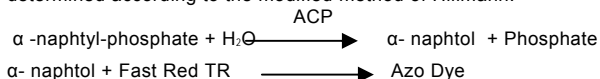




**Acid phosphatase
α-Naphtyl phosphate. Kinetic
Quantitative determination of acid phosphatase
(ACP)
Store at 2-8°C**

PRINCIPLE OF THE METHOD

Hillmann method: acid Phosphatase activity present in the sample is determined according to the modified method of Hillmann.



α-naphtol reacts with a diazoted compound forming a colour with a maximum of absorbance at 405 nm. Tartrate is used as specific of the prostatic fraction.

CLINICAL SIGNIFICANCE

Acid phosphatase is an enzyme present in almost all weaves of the organism, being particularly high in prostate, stomach, liver, muscle, spleen, erythrocytes and platelets. High levels of acid phosphatase are found in prostatic phatologies as hypertrophy, prostatitis or carcinoma. In hematological disorders, bones or liver diseases as well as in Paget's or Gaucher's diseases. Decreased serum acid phosphatase has no clinical significance^{1,9} Clinical diagnosis should not be made on a single test result; it should integrate clinical and other laboratory data.

REAGENTS

R1 Buffer	Sodium citrate pH 5.2	50 mmol/L
R 2 Substrate	α-Naphtyl phosphate Fast Red TR	10 mmol/L 6 mmol/L

PREPARATION

Working reagent (WR):
Dissolve one tablet of R 2 Substrate in 15ml of R 1 Buffer.
Cap and mix gently to dissolve contents.
Stability: 2 days at 2-8°C or 6 hours at room temperature.

STORAGE AND STABILITY

All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8°C, protected from light and contaminations prevented during their use.
Do not use the tablets if appears broken.
Do not use reagents over the expiration date.

Signs of reagent deterioration:

- Presence of particles and turbidity.
- Blank absorbance (A) at 450 nm > 0,44.

ADDITIONAL EQUIPMENT

- Spectrophotometer or colorimeter measuring at 405 nm.
- Thermostatic bath at 30°C o 37°C (± 0.1°C)
- Matched cuvettes 1.0 cm light path.
- General laboratory equipment.

SAMPLES

Serum. Use only clear and unhemolyzed serum, separated from the clot as soon as possible. Do not use plasma.

PROCEDURE

1. Assay conditions:
Wavelength:405 nm
Cuvette :1 cm light path
Constant temperature 30°C / 37°C
2. Adjust the instrument to zero with distilled water or air.
3. Pipette into a cuvette:

	ACP Total (T)
WR (mL)	1.0
Sample (µL)	100

4. Mix, incubate for 5 minute.
5. Read initial absorbance (A) of the sample, start the stopwatch and read absorbance at 1 minute intervals thereafter for 3 minutes.
6. Calculate the difference between absorbance and the average absorbance differences per minute (ΔA/min).

CALCULATIONS

ΔA/min x 750 = U/L of ACP (T)
750 x (ΔE/min ACP (T) - ΔE/min ACP Non inhibitor by Tartrate) = U/L of ACP prostatic.

Units: One international unit (IU) is the amount of enzyme that transforms 1 µmol of substrate per minute, in standard conditions. The concentration is expressed in units per litre of sample (U/L).

QUALITY CONTROL

Control sera are recommended to monitor the performance of assay procedures. If control values are found outside the defined range, check the instrument, reagents and technique for problems. Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

REFERENCE VALUES

	30°C	37°C
Total acid phosphatase:		
Men:	< 4.3 U/L	< 5.4 U/L
Women:	< 3.1 U/L	< 4.2 U/L
Prostatic acid phosphatase	< 1.5 U/L	< 1.7 U/L

These values are for orientation purpose; each laboratory should establish its own reference range.

PERFORMANCE CHARACTERISTICS (Total ACP)

Measuring range: From detection limit of 0,13 U/L to linearity limit of 150 U/L. If the results obtained were greater than linearity limit, dilute the sample 1/2 with NaCl 9 g/L and multiply the result by 2.

Precision:

	Intra-assay (n=20)		Inter-assay (n=20)	
Mean (U/L)	23.67	2.56	23.6	2.6
SD	0.22	0.07	0.22	0.07
CV (%)	0.95	2.90	0.92	2.76

Sensitivity: 1 U/L = 0.0034 A/min.

Accuracy: Results obtained using Atlas reagents did not show systematic differences when compared with other commercial reagents. The results obtained using 50 samples were the following:

Correlation coefficient (r): 0.99

Regression equation: y= 0.9977x + 0.1486.

The results of the performance characteristics depend on the analyzer used.

INTERFERENCES

Hemolysis interferes due the high concentration of acid phosphatase in red cells. A list of drugs and other interfering substances with acid phosphatase determination has been reported by Young et al.

BIBLIOGRAPHY

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