ACP
(Colorimetric Test with α-Naphthylphosphate)

Cat.No	Package Size
102 080	5 x 10 ml R1/ 50 ml BufferR2 / 50 ml Buffer-Tartrate R3 / 2 ml Stabilizer R4

METHOD / REACTION PRINCIPLE:
α-Naphthylphosphate is hydrolysed by ACP to phosphate and α-naphthol, which is converted with FRTR-salt into an azo dye. The increase of absorbance at 405 nm is proportional to the total ACP activity in the sample. The prostatic acid phosphatase (PACP) can be blocked by tartrate and can be determined indirectly (through the non-prostatic ACP) by calculation of the activity difference.

α-Naphthylphosphate + H_20 —ACP——>
phosphate + 1-naphthol
α-Naphthol + FRTR-salt ————————> Azo dye

REAGENTS: (Concentrations in the test)

R1 : 1-Naphthyl phosphate	10 mmol/l
Fast Red TR-salt (4-chloro-2-methylphenyl diazonium salt)	1.5 mmol/l
R2 : Citrate buffer pH 5.2	100 mmol/l
R3 : Citrate buffer pH 5.2	100 mmol/l
Tartrate	135 mmol/l
R4 : Stabilizer = Acetic acid	0.8 mol/l

The sealed reagents are stable up to the indicated expiry date if stored at 2° - 8°C.

PREPARATION AND STABILITY OF WORKING REAGENTS

R2 and R3 are ready for use

Reagent A (determination of **T**otal **ACP**):
Dissolve the contents of R1 (substrate) in 10 ml of buffer solution R2.
Mark label with **„A".**
Reagent B (determination of **P**rostatic **ACP**):
Dissolve the contents of R1 (substrate) in 10 ml of tartrate solution R3. Mark label with **„B".**

Stability of Working reagents
3 days at 2° - 8°C
1 day at 18° - 25°C

SAMPLES :
Serum, no plasma! Avoid hemolysis!
Use immediately or stabilize :
Add 1 drop of 0.1% acetic acid to 1 ml of serum:
ACP is stable for 3 days at 2-8°C .

NORMAL RANGES

Total Acid Phosphatase

Men	≤ 4,7 U/l
Women	≤ 3,7 U/l

Prostatic Acid Phosphatase
≤ 1,6 U/l

ASSAY PROCEDURE:
Wavelength : 405 nm Hg
Light path: 1 cm
Temperature : 37 °C
Measurement: against air (increasing absorbance)

Pipette into cuvettes:	A (TACP)	B (NPACP)
Sample	100 µl	100 µl
Reagent A	1000 µl	-
Reagent B	-	1000 µl

Mix, read absorbance A_1 after 5 min and start stop-watch at same time. Read absorbance A_2 exactly after 3 min => Calculate ΔA/min.

CALCULATION:
Calculate total acid phosphatase activity and prostatic acid phosphatase activity using following factors :

Total acid phosphatase ***TACP (U/l) = ΔA/min x 743***
Non-prost. acid phosph. ***NPACP (U/l) = ΔA/min x 743***

Prost. acid phosph. ***PACP (U/l) = TACP - NPACP***

Conversion factor
traditional units (U/l) into SI-units (µkat/l):
1 U/l = 16.67 x 10^{-3} µkat/l

CALIBRATORS AND CONTROLS
For the calibration of automated analyzers Greiner Multicalibrator is recommended, for quality control use Greiner normal and abnormal control, Unitrol I and Unitrol II.

LINEARITY:
If absorbance change exceeds 0.3 at 37°C, or if the activity is higher than 74 U/l, dilute 0.1 ml of sample with 0.2 ml phys. saline (0.9 %) and repeat the assay using dilution. Multiply result by 3.

LITERATURE

1. Thomas L ed. Clinical Laboratory Diagnostics.
2. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft, 1988:147
3. Hillmann G. Z.Klin.Chem.Klin.Biochem.9, p.273.

03/2010

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