CHLORIDE

(Thiocyanate) End-Point

BIO LAB DIAGNOSTICS
ISO 9001:2008
ISO 13485:2012
CE

CLINICAL SIGNIFICANCE

Low Chloride levels in blood are associated with gastrointestinal or renal salt losing conditions or metabolic acidosis and Addison's disease. Increased levels are observed in cases of dehydration, renal obstruction, nephritis, congestive heart failure and Cushing's syndrome.

PRINCIPLE

$2Cl^- + Hg^+(SCN)^{-2} \longrightarrow HgCl_2 + (SCN)$

$2(SCN^-) + 2Fe^{++} \longrightarrow 2Fe^{++}(SCN^-)$

The formation of **REDDISH BROWN COLOURED** ferric thiocyanate can be measured at 460nM and is directly proportional to Chloride concentration.

REAGENTS COMPOSITION

1. CHLORIDE REAGENT	**2x50 mL**
Ferric Nitrate	100 mMol/L
Mercuric Thiocyanate	10 mMol/L
Methanol	3 Mol/L
Contains preservatives and stabilizers.	
2. CHLORIDE STANDARD	**3 mL**
Sodium Chloride	100 mMol/L
Contains surfactants and stabilisers.	

Working Reagent Preparation

All reagents are ready to use.

STORAGE AND STABILITY

When stored at R.T. and protected from light, the reagents are stable until the expiry date stated on the label.

SAMPLE

Specimen can be unhaemolysed serum, heparinised plasma, CSF, body fluids or urine. Urine sample to be diluted 1 to 2 with glass distilled water.

SYSTEM PARAMETERS

Reaction	End-point
Temperature	25°C to 35°C (RT)
Wavelength	460 nM (440-500 nM)
Absorbance Range	1Å
Cuvette Path Length	1 cM

Reagent Volume	**500 µL**	**1000 µL**
Sample Volume	**10 µL**	**10 µL**

Reaction Time	5 Mins
Dynamic Range	70-140 mMol/L
Max. limit of Blank Reagent	0.2000
Final Colour Stability	30 Mins.

MANUAL PROCEDURE

1. Wash all glasswares, pippette tips and pipettes thoroughly with glass distilled water. DRY.

2. **Pipette into 3 test tubes**

	Blank mL	Standard mL	Test mL
Chloride Colour Reagent 1	3.00	3.00	3.00
Distilled Water	0.02	-	-
Standard Reagent No. 2	-	0.02	-
Sample	-	-	0.02

Mix well. Incubate for 5 minutes at R.T. Read Optical Density (OD) at 460 nM (440-500nM) or BLUE filter against Blank. Final colour is stable for 30 minutes away from bright light.

NOTE: Reagent and sample volume can be altered proportionately.

MICRO PROCEDURE FOR DISCRETE ANALYSERS

Cuvette Volume	**1 mL**	**0.5 mL**
Chloride Colour Reagent	1000 µL	500 µL
Sample or Standard	10 µL	10 µL

Mix well. Incubate at R. T. for 5 mins. Read OD at 460 nM (440-500 nM) against Blank. The final colour is stable for 30 minutes away from bright light.

Note: Programme the analyser using system parameters. A specific data sheet may be provided for each analyser on request.

RESULTS

$$\text{Chloride in mMol/L} = \frac{\text{OD Test}}{\text{OD STD}} \times 100$$

$$\text{in mEq/dL} = \frac{\text{O.D. Test - O.D. Blank}}{\text{O.D. STD - O.D.Blank}} \times 100$$

mGs/dL = mEq/L x 3.545

EXPECTED VALUES

Serum	96	-	106 mEq/L
	340	-	375.8 mGs of Chlorine/dL
	561	-	619.4 mGs of NaCl/dL
CSF	123	-	135 mEq/L
	436	-	478.6 mGs of Chlorine/dL
	728.8	-	788.9 mGs of NaCl/dL
Urine	170	-	250 mEq / 24 hours
	602.7	-	886.3 mGs of Chlorine / 24hrs
	993.5	-	1461 mGs of NaCl / 24hrs

Values vary subject to the infusion of sodium chloride. As with all diagnostic methods, the final diagnosis should not be made on the result of a single test as well as laboratory diagnosis must be confirmed with clinical manifestations.

LIMITATIONS

This reagent system is highly sensitive to chloride radicals commonly found as contaminants in tap water and detergents etc. Test tubes and glasswares washed in tap-water may lead to false results. ***This methodology is not useful to measure outside dynamic range of 70-140 mMol/L.*** For levels out side the dynamic range of methodology increase or decrease sample volume and multiply the result appropriately. For more accuracy each kit should be tested against a set of standards with different concentrations and a graph be plotted.

WARNING

This reagent system is for ***in vitro*** use only.
This reagent system is containing preservatives and components that have not established for safety if contacted on broken skin or eye or taken orally. In case of such incidents wash off with plenty of water, or consult a physician.

QUALITY CONTROL

To ensure adequate quality control, each kit should be tested against a standard control sera. It should be realised that the use of quality control material checks both instrument and reagent function together. Factors which might affect the performance of this test include proper instrument function, temperature control, cleanliness of glasswares and accuracy of pipetting.
It is appropriate to establish each laboratory's accuracy constant and interpret values accordingly. Similarly, laboratory findings should be established by clinical manifestations.

BIBLIOGRAPHY

1. SHOENFELD, R.G.Clin. Chem. 10,533.
2. ZALL, D.M. et. al. Anal. chem. 28, 1665, 1956.

BIOLAB DIAGNOSTICS (I) PVT. LTD.
J-245, MIDC, TARAPUR, BOISAR - 401 501, MAHARASHTRA.
E-mail : biolab@vsnl.com / www.biolabdiagnostics.com
Customer Care : (+ 9122) 28088243