CHLORIDE CE

CONTENTS			
REF	1116005	Chloride	2 x 50 mL
For *in vitro* diagnostic use only			

CHLORIDE

THIOCYANATE

Colorimetric method

ENDPOINT

PRINCIPLE

Chloride ions in the quantitatively displaces thiocyanate from mercuric thiocyanate. Liberated thiocyanate ion reacts with ferric ion forming a red ferric-thiocyanate complex proportional to the concentration of chloride present in the sample.[1,2]

$$2\ Cl^- + Hg(SCN)_2 \longrightarrow HgCl_2 + 2\ (SCN)^-$$

$$3\ (SCN)^- + Fe^{3+} \longrightarrow Fe(SCN)_3$$

REAGENT COMPOSITION

R1 **Thiocyanate reagent**. Mercuric thiocyanate 2 mmol/L, mercuric nitrate 0.1 mmol/L, iron nitrate 30 mmol/L, HNO_3 45 mmol/L. (see Notes). $\mathbf{X_n}$

CAL **Chloride standard.** Chloride 125 mEq/L (mmol/L). Organic matrix based primary standard.

STORAGE AND STABILITY

Store at 15-30ºC.
The Reagents are stable until the expiry date stated on the label.

REAGENT PREPARATION

The Reagent and Standard are ready-to-use.

SAMPLES

Serum, heparinized plasma and CSF.

The sera are stable in capped tubes for at least 4 hours at room temperature, 2 days refrigerated (4-8ºC) and several months frozen (-20ºC).
Cerebrospinal fluid should be collected in three sterile tubes, with samples from the first or second tube used for chloride determinations.

INTERFERENCES

- Bilirubin (>12 mg/dL), albumin (>15 g/L) and triglycerides (6 g/L), do not alter the assay.[2]
- EDTA and fluoride anticoagulants will interfere with the assay.
- Bromide ions present in the sample from therapeutic use cannot be distinguished from chloride. Since they contribute a total of less than 1.0 mmol/L of serum, their presence can be neglected.

MATERIALS REQUIRED

- Photometer or colorimeter capable of measuring absorbance at 470 ± 10 nm.
- Constant temperature incubator set at 37ºC (optional).
- Pipettes with disposable plastic tips to measure reagents and samples.
- Disposable plastic tubes for the tests.

PROCEDURE

1. Bring reagents and samples to room temperature.
2. Pipette into labelled test tubes:

TUBES	Blank	Sample	Standard
Reagent	1.0 mL	1.0 mL	1.0 mL
Sample	–	10 µL	–
Standard	–	–	10 µL

3. Mix gently by inversion one or two times. *Do not shake or stir vigourously.*
4. Incubate the mixture for 5 or 10 minutes at a selected constant temperature between 25-37ºC (see Notes).
5. Read the absorbance (A) of the samples and the standard at 470 ± 10 nm against the reagent blank.

The color is stable for about 2 hours, at room temperature, protected from light.

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CALCULATIONS

Serum, plasma

$$\frac{A_{Sample}}{A_{Standard}} \times C_{Standard} = \text{mEq/L (mmol/L) chloride}$$

Samples with concentrations higher than 125 mEq/L (125 mmol/L) should be diluted 1:2 with distilled water and assayed again. Multiply the results by 2.
Concentrations less than 75 mmol/L, should be brought within linear range by increasing the amount of serum used.

REFERENCE VALUES[3]

Serum, plasma	98-111 mEq/L (98-111 mmol/L)
CSF	120-130 mEq/L (120-130 mmol/L)

It is recommended that each laboratory establishes its own reference range.

QUALITY CONTROL

The use of a standard to calculate results allows to obtain an accuracy independent of the system or instrument used.
To ensure adequate quality control (QC), each run should include a set of controls (normal and abnormal) with assayed values handled as unknowns.

REF **1980005** HUMAN MULTISERA NORMAL
Borderline level of chloride. Assayed.

REF **1985005** HUMAN MULTISERA ABNORMAL
Elevated level of chloride. Assayed.

CLINICAL SIGNIFICANCE

Sodium and chloride represent the majority of the osmotically active constituents of plasma. As a result, chloride is significantly involved in maintenance of water distribution, osmotic pressure, and anion-cation balance in the extracellular fluid compartment.

Hypochloremia (decreased plasma Cl^- concentration) is observed in salt-losing nephritis as associated with chronic pyelonephritis. In Addison´s disease, Cl^- levels as well as Na^+ levels may drop significantly during the Addisonian crisis and in certain types of metabolic acidosis (e.g., diabetic ketoacidosis and renal failure) and aldosteronism. In metabolic alkalisis, plasma levels of Cl^- tend to fall while HCO_3^- levels increase.

Hyperchloremia (increased plasma Cl^- concentration) occurs with dehydration, renal tubular acidosis, diabetes insipidus, acute renal failure, adrenocortical hyperfunction and metabolic acidosis. Extremely high dietary in take of salt and overtreatment with saline solutions are also causes of hyperchloremia.

ANALYTICAL PERFORMANCE

- ***Linearity.*** Between 70 and 125 mmol/L
- ***Precision***

mmol/L	Within-run			Between-run		
Mean	75	100	120	75	101	121
SD	1.3	2.2	1.7	1.8	2.5	1.7
CV%	1.7	2.2	1.4	2.4	2.5	1.4
N	10	10	10	10	10	10

Replicates: 10 for each level. Replicates: 10 for each level for 8 days.
Instrument: CECIL CE 2021

- ***Sensitivity.*** Using a 1:100 sample/reagent at 470 nm, 1mmol/L of chloride will produce a net absorbance of approximately 0.005.
- ***Correlation.*** This assay (y) was compared with a similar commercial method (x). The results were:
N = 20 r = 0.993 y = 1.05x – 3.1

NOTES

- The reagent is harmful if swallowed. **Do not pipette by mouth**. In case of accident or if you feel unwell, seek medical advice immediately.
- A 5-min incubation period is satisfactory when the incubation and assay are performed at 37ºC. A 10-min incubation period is recommended when the incubation and assay are performed at room temperature.
- For optimum results, the glassware used for the chloride procedure should be scrupulously clean. It may be found convenient to acid wash the material (H_2SO_4-$K_2Cr_2O_7$) and then thoroughly rinse it with distilled water.

REFERENCES

1. Schoenfeld R.G., and Lewellen Clin. Chem. 10 : 533 (1964).
2. Tietz. N.W. Clinical Guide to Laboratory Tests, 3rd Edition. W.B. Saunders Co. Philadelphia, PA. (1995).
3. Young, D.S. Effects of Drugs on Clinical Laboratory Tests. 4 th Edition. AACC Press (1995).

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